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Indianapolis, Indiana 46204

EXECUTIVE COMMITTEE, COUNCIL AND BUDGET COMMITTEE

Executive Committee

*Otto K. Behrens
Alice S. Bennett
William Beranek, Jr.
Warren William Bowden
Ken Brunson
Lois Burton
Ernest E. Campaigne
Margaret S. Cavanaugh
Richard L. Conklin
*Donald J. Cook
Robert H. Cooper
Walter A. Cory, Jr.
Theodore J. Crovello
Fay K. Daily
*William A. Daily
*Harry G. Day
*Clarence F. Dineen
William N. Doemel
Douglas Duff
*William R. Eberly
*Raymond E. Girton
*Arthur T. Guard
*Frank A. Guthrie
*Robert E. Henderson
Donald R. Hendricks
*William B. Hopp
Marion T. Jackson
*Willis H. Johnson
Duvall A. Jones
Gene Kritsky
*Alton A. Lindsey
*C. A. Markle
John H. Meiser
*Melvin C. Mellon
*A. H. Meyer
*Howard H. Michaud
Benjamin Moulton
Craig Nelson
*Jerry J. Nisbet
*John B. Patton
Robert R. Pinger
*S. N. Postlethwaite
*H. M. Powell
Roger Roth
*Damian V. Schmelz
David M. Sever
Akhtar H. Siddiqi
John T. Snow
David F. Spencer
Patrick J. Sullivan
Curtis H. Tomak
*W. J. Wayne
*J. Dan Webster
*Winona H. Welch
Kathryn J. Wilson
Donald R. Winslow
*Howard R. Youse
Patricia A. Zeck

*Past President of Academy

Council

President—William R. Eberly
President-Elect—Alice S. Bennett
Secretary—John Meiser
Treasurer—Duvall A. Jones
Director of Public Relations—Walter A. Cory, Jr.
Editor—Donald R. Winslow
Chairman, Science & Society Committee—Craig Nelson

Budget Committee

President—	William R. Eberly
President-Elect—	Alice S. Bennett
Immediate Past President—	Robert E. Henderson
Secretary—	John H. Meiser
Treasurer—	Duvall A. Jones
Director of Public Relations—	Walter A. Cory, Jr.
Editor—	Donald R. Winslow
Chairman, Science & Society Committee—	Craig Nelson
Co-Director, Junior Academy of Science—	Keith Hunnings
Co-Director, Junior Academy of Science—	Leota Skirvin
Chairman, Program Committee—	Theodore J. Crovello
Chairman, Library Committee—	Lois Burton
Chairman, Youth Activities Committee—	Jerry J. Nisbet
Chairman, Academy Foundation—	William A. Daily
Chairman, Research Grants Committee—	Ernest E. Campaigne

Indiana Academy of Science
SPRING MEETING
MINUTES OF THE EXECUTIVE COMMITTEE MEETING
April 23, 1982

President William R. Eberly called the meeting to order at 4:13 p.m. in Room 202, Center for Continuing Education at the University of Notre Dame, South Bend, Indiana.

TREASURER'S REPORT

The Academy finances as of April 21, 1981 were presented by Treasurer Duvall A. Jones.

Current Assets

Checking Account	$ 6,105.83*
Savings Account	$16,323.51
Total	$22,429.34

*Check for $990 not yet negotiated

These are distributed in

Academy Accounts	$ 9,203.47
Administered Accounts	$12,235.87
Total	$21,439.34

Acceptance of the report was moved, seconded, and approved.

ELECTED COMMITTEE REPORTS

Academy Foundation Committee

William A. Daily, Chairman, reported that on March 31, 1982 the John W. Wright Invested Income Account held $65,000 in two Commercial Paper Funds, yielding an average of 15.43%. An additional $30,000 is held in U.S. Treasury Notes 15 1/8%. An additional $6,000 was in cash to be invested in Commercial Paper.

The Foundation account was $27,712, and the market value of the John S. Wright Fund was $605,999.

Research Grants Committee

Walter A. Cory, Jr. presented the committee report for E. Campaigne, Chairman. Total research applications numbered 25 with requests for $22,000, about 100% more than usual. Generally the Spring awards are about 60% of requested funds. This would require $13,000 if the previous practice is followed, leaving $2,000 for the Fall round.

Motion: To make a maximum of $21,000 available for research grants using $6,000 appropriated from last year but left unspent. Seconded. The motion passed unanimously.

Campaigne also requested a clarification of Academy support for Secondary School grants for the summer. In 1980, AAAS supplied $600 supplemented by $90 of Academy funds to award eight Secondary School Grants. In 1981, $300 was received from AAAS and was supplemented by $298 from Academy funds to award six grants.

President Eberly pointed out that funds have been appropriated for the purpose of giving grants in case AAAS funds become unavailable.

COMMITTEE REPORTS

Academy Representative to NAAS

John H. Meiser, appointed representative of the Academy to attend the Washington AAAS meeting, reported that due to certain circumstances he was unable to attend the meeting.

Youth Activities Committee

John H. Meiser presented the report for Jerry J. Nisbet, Chairman. Leota Skirvin, Director of the Junior Academy has confirmed facility arrangements for the autumn meeting of the Junior Academy at the University of Notre Dame.

Karl Kaufman and Lloyd Anderson, Co-Directors of Indiana Science Fairs report financial support for five $1,200 scholarships and for a financial subsidy of approximately $21,900 to defray travel expenses for the Science Fair Winners to attend the International Science Fair will be provided by The Indiana Science Education Fund, Inc.

Also reported from Walter Cory, Jr., Director of the Indiana Science Talent Search, was that 49 students entered competition for the state talent search this year. Of this number, 26 were selected as finalists with two of these, Richard Meyer, East Noble High School and Lynn McCaslin, R. N. Snyder High School, awarded $1000 scholarships. Kappa Kappa Kappa has continued its financial support of the Indiana Science Talent Search program.

A meeting of the Youth Activities Committee was held on February 2, 1982 with reports and discussions on each of the Academy's Youth Activities programs including ways of making more science students aware of the Academy's grant programs for research support.

Jerry Colglazier had sent a report that two of the 25 national finalists and one of the two national winners of the Thomas Edison/McGraw Scholarship Program were Indiana students. Thomas Beutner, Marquette High School, Michigan City, is the Indiana winner.

It was pointed out from the floor that a number of state winners in these programs are Academy members. Additional chapters of the Junior Academy are to be encouraged through personal contact.

Library Committee

President William R. Eberly reported for Lois Burton, Chairwoman. Proceedings for 1980 have been sent. Eberly raised the question of the future of the Academy library now housed in State Library building. No conclusions were reached.

Program Committee

Theodore J. Crovello, Chairman, indicated that there were 28 paid dinners from Indiana Academy members and 60 from Sigma Xi members for the evening's joint dinner meeting. This meeting marks the first time IAS has met with the Northern Indiana Natural Areas Conference.

The Fall meeting will accommodate the Junior Academy at Stephen Center. The question of a registration fee for meetings was raised. The question was discussed and the suggestion was made to handle each meeting on an individual basis through the Invitations Committee. The question of having organizations, vendors, and schools setting up booths at our meetings was discussed. This is important because the symposium on Saturday will be Computers and Education. Should there be charges, and how much? By consensus, there is support for exhibitors setting up booths. Crovello is to investigate the possibility of charging fees for space. The Saturday program will have two sessions: the first focusing on computers in science education, the second on value oriented science education.

Publication Committee

President Eberly gave the report for Benjamin Moulton that several monographs are in preparation and one should be off the press soon.

Donald R. Winslow, Editor, reported that there is no longer a master contract by the State for printing the Proceedings. Art Craft Press, Inc. will print Vol. 91 whereas Western Newspaper Publishing Co., Inc., published Vol. 90 on an offset press and on white book paper rather than by hot lead and on an enamel printing stock (the way it has previously been printed).

The editing has been done on Vol. 91. Some 75 papers were reviewed and 70 were recommended for publication.

Science and Society

It was reported that there would be a meeting on May 7th in Indianapolis.

Membership Committee

Alice S. Bennett, Chairperson, noted the need for new members and indicated the best way to attract new members is through personal contact, particularly with faculty and people in industry.

Invitations Committee

President Eberly gave the report for Donald Cook, Chairman. The 1983 meeting will be held at the I.U. Purdue Campus at Fort Wayne. Dr. Haddock may be program chairman. The 1984 meeting will be held at Butler University.

Emeritus Membership Selection Committee

John H. Meiser presented two names in nomination for Dr. Robert H. Cooper, Chairman: Mrs. George F. Martin, Membership 1951 through 1981 and was a division chairman; Dr. Robert L. Stone, Membership 1955 through 1981. It was moved and seconded to make the nominees emeritus members. The motion passed.

Biological Survey Committee

Theodore J. Crovello, BSC Chairman listed six items: 1. Questionnaires about publications or works in progress concerning biota or environment were distributed. 2. Selected results from returned questionnaires will appear in the Proceedings. 3. Data from each questionnaire is being entered into the computer data bank. 4. A proposal has been submitted to publish approximately 3,000 references of the cumulative bibliography. 5. The contract from the U.S. Department of Interior to study rare plants in the State was completed. 6. The BSC and Academy are cosponsoring at this Spring meeting, The Northern Indiana Natural Areas Conference and Field Day with over 140 professionals in attendance.

NEW BUSINESS

Constitution Revisions

Eberly led a discussion of some changes to be considered to bring our normal operations in line with the constitution, such as having the Editor be chairman of the Publications Committee. Another area of concern discussed was the relation of the Academy to the State.

Symposium on Indiana-Kentucky Boundary

This question is now referred to the Science and Society Committee.

Academy Council Functions

Eberly discussed the possibility of having the Academy Council meet with

the program chairman to oversee the entire meeting arrangement process. No decision was made.

The meeting adjourned at 5:26 p.m.

The meeting was reconvened at 9:50 p.m. This was to have been the General Meeting. However, due to the lateness of the hour and because mostly only the previous attendees were present, it was decided to continue in executive session.

President Eberly discussed the possibility of speaking on Creationist Beliefs for his address to the Academy. The Resolution on Teaching Creationist Beliefs in Public School (AAAS) was referred to the Science and Society Committee.

Discussed at some length was the mailings to members, especially the Call for Papers. It was moved and seconded to send all calls for papers in a general mailing. Passed. This will be tried in the Newsletter in order to hold down the cost of duplication of individual mailings.

The meeting adjourned at 10:21 p.m.

Indiana Academy of Science
FALL MEETING
MINUTES OF THE EXECUTIVE COMMITTEE MEETING
October 28, 1982

President William R. Eberly called the meeting to order at 7:15 p.m. in Room 202, Center for Continuing Education at the University of Notre Dame, South Bend, Indiana.

SECRETARY'S REPORT

Secretary John H. Meiser read the minutes of the Executive Committee Meeting of April 23, 1982. The minutes were approved as amended.

TREASURER'S REPORT

Treasurer Duvall A. Jones presented the Academy finances as of October 27, 1982.

Current Assets	
Checking Accounts	$ 4,457.54
Savings Accounts	17,328.50
	$21,786.04
These are distributed in	
Academy Accounts	$10,698.16
Administered Accounts	11,087.88
Total	$21,786.04

Membership totals on October 27, 1982 were 987 consisting of 31 sustaining, 533 senior, 243 regular, 97 emeritus and the rest in other categories. It was moved and seconded to accept the treasurer's report. The motion passed. Jones reported that some inconsequential invoices and other paper work more than three years old would be discarded due to lack of space for storage.

EDITOR'S REPORT

Donald R. Winslow, editor, reported some of the problems with the production of *Vol. 91* of the *Proceedings* including the faulty typesetting and the possible problem of funding restrictions by the State. It now appears that *Vol. 91* will be ready sometime in December.

DIRECTOR OF PUBLIC RELATIONS

Walter A. Cory, Director, discussed the use of the differently colored name tags and publicity for the meeting.

PROGRAM COMMITTEE

Theodore J. Crovello, Chairman, distributed a sheet listing last minute changes in the announced program. He also indicated that papers for this meeting were down slightly from last year, 187 against 204 last year.

REPORTS, ELECTED COMMITTEES

Academy Foundation Committee

William A. Daily, Chairman, reported that on September 30, 1982 that the Foundation account had a market value of $31,218.78 and had earned $2,421.86 in the last year. The John S. Wright Fund had income of $35,255.37 from 10-01-81 to 9-30-82 of which $27,724.39 was transfered to the Invested Income Account. The Fund had a market value of $581,004.32 and fluctuates with the value of the investment portfolio. The Invested Income Account earned $11,324.40 in the same period and had a net value of $104,217.28 on 9-30-82. Money is transfered from this latter account to support Academy publications and research grants.

Bonding Committee

Duvall A. Jones reported for Frank Guthrie that the old Faternal Order Form bond has been changed to a Position Schedule Form bond to cover the person in the office of treasurer.

Research Grants Committee

Ernest Campaigne, Chairman, reported that for the Fall round of grants, 12 proposals had been received with 4 fully funded and 7 partially funded spread over 8 institutions. Some $7,659 were awarded with the average being $696. Nine secondary school awards were granted for a total of $840. A considerable increase in both the number of applications and the dollar amounts was noted. The awards now follow.

Investigator/Institution Spring, 1982	Abbreviated Title	Amount
Berquist, A.M. Notre Dame	Zooplankton Assemblages in Dune Ponds	$685
Brown, R. E., Dr. Huntington College	Survey of Flora and Fauna at Salamonie Reservoir	$536
Cave, J.B. Ball State	Fish Mortality/Angling Gear	$183
Fogle, T.A. St. Mary's College	Cytogenetic Analysis of Zoo Monkeys	$600
Greene, R. W., Dr. Notre Dame	Symbiotic Association of Flatworm and Algae	$850
Greenlee, J. K. Notre Dame	Genetic and Morphological Differentiation	$525
Hogarth, A.J.C.L. DePauw	Redox Properties of Cu(II)perchlorate	$850
Hughes, A.L. Indiana University	Seasonal Trends in Body Size of Mosquitofish	$185

Investigator/Institution	Abbreviated Title	Amount
Kamo, K. Indiana University	Alkaloid Biosynthesis in Opium Poppy	$625
Krohne, D.T. Wabash	Structure of Populations of *Peromuscus leucopus*	$735
Litton, J.R. St. Mary's College	A New Vitamin E Compound	$500
Mandracchia, M.J. Notre Dame	Glochidium larva	$725
Mays, C.D. DePauw	Murine Reproduction, Effects of Smoke On	$500
McCreary, N.J. Notre Dame	Competition and Coexistence in Freshwater Perennials	$860
Mould, E. D. St. Mary's College	Effect of Phenolics on Forage of Herbivores	$655
Riemenschneider, V.L. IUSB	Wind Damage Survey (Renewal)	$105
Schwarzwalder, R. Indiana University	Analysis of Platanaceae of Mexico	$650
Stewart, C.A. Indiana University	Cycles of Lipids in Eastern Gray Squirrels	$575
Swartz B.K., Dr. Ball State	Radiocarbon Dating of Bertsch Site, Wayne Co.	$185
Wasser, J.S. Indiana University	Hibernation in Northern Water Snake	$775
	Total Recommended	$11,304

Fall, 1982

Carter, C. Purdue University	Effects of Concepts and Strategies on Learning	$616
Dhawale, S.W. Indiana Uni. East	Stability of Sodium Thiosulfate in Acid Solutions	$485
Vas Dev Notre Dame	Genetics of Speciation in *Aedes* (*Stegomyia*) *scutellaris*	$470
Huang, C.T. Purdue University	Feasibility of Long Wall Coal Mining in Shallow Overburden, Warrick Co.	$520
Latino, J.C. Notre Dame	Role of Zooplankton in Nutrient Cycling in Dune Ponds	$895
MacMillan, P.C. Hanover College	Analysis of Log Samples from Michigan and Pennsylvania	$1040
Rybarczyk, J.P. Ball State	Effect of Sample Particle Size and Composition on Excitation Mechanisms in Inductively Coupled Plasma	$1000

Investigator/Institution	Abbreviated Title	Amount
Sever, D.M. St. Mary's College	Cold-blooded Vertebrates of Kingsbury State Fish and Wildlife Area	$683
Vaughn, M. Indiana State	Calcium Uptake in Subcellular Membrane from Corn	$725
Weeks, H.P., Jr. Purdue University	Regulatory Effects of Supplementary Sodium on Wild *Microtus pennsylvanicus*	$940
Yoder, S.C. Goshen College	Tree Survey in Proposed Addition to Merry Lea State Nature Reserve	$285
	Total Recommended	$7659

Secondary School Research Grants

Name/School/Sponsor	Research	Amount
Alan Engel John Adams H.S., South Bend Nevin Longenecker	Possible Effects of Altered Diets on Allergic Reactions	$100
James A. Barsic Marquette H.S., Michigan City Dean G. Christakis	Scanning Electron Microscope Studies of Metallic Stress Fractures	$75
Susan Rucinski Gavit H.S., Hammond M. Kobe	Formation and Movement of Sand Dunes vs Increasing Amounts of Industrial Pollution	$100
Tamara M. Hughes Plainfield H.S., Plainfield Mrs. Nell Walthers	Effects of Low pH on Bluegills and Catfish	$100
Valori Miller, Jeff Medwetz, Scott Bearby Gavit H.S., Hammond M. Kobe	Vitamins as Plant Growth Stimulants	$115
Jim Szalony Gavit H.S., Hammond M. Kobe	Effects of Contaminated Flood Water on Soil Microfauna	$100
Laura Warot Gavit H.S., Hammond M. Kobe	Effect of Highway Salt Runoff on Adjacent Ponds	$100
Steve Longenecker Riley H.S., South Bend John Koellner	Relative Rates of Absorption of *Alpha* and *Beta*-glucose	$100
Kathleen Ramos Gavit H.S., Hammond M. Kobe	Effect of Tobacco and Steel mill Smoke on Bacteria	$50
	Total Approved	$840

REPORTS, APPOINTED COMMITTEES

Youth Activities Committee

Leota Skirvin, director of the Junior Academy, announced that this was the 50th year of the Junior Academy; the first meeting was here at Notre Dame. Concern was voiced that this year the Junior Academy coincides with the annual State Conference on Instruction for teachers. It appears that some teachers will not be able to participate. However, this might be remedied next year by having the Junior Academy included on the list of approved events in the Conference on Instruction. Other concerns will be discussed at the Junior Academy Council in February.

Walter A. Cory reported that work for the Science Talent Search on December 15 is progressing nicely and he expects 40-50 to participate.

Reporting for Karl L. Kaufman, director of Youth Science Development, Cory reported that of the 9 one-day workshops for Fall 82, 7 have been held with approximately 100 people attending each. The regional fairs are in March and April again with the national meeting in Albuquerque. A note of appreciation was voiced for all the work done by Karl Kaufman.

Library Committee

Lois Burton, Chairman, reported that the *Proceedings Vol. 90* has been distributed to some 500 libraries, institutions, and societies. The "Requisition for Printing" for *Vol. 91* was submitted to the Indiana Department of Administration and the Budget Agency in August. In addition 108 volumes were prepared for binding and 107 interlibrary loan requests were filled during the year.

Nomination Committee

Nominations forwarded by Robert Henderson, chairman, were presented by Cory.

President-elect	Theodore J. Crovello
Secretary	Richard Conklin
Research grants	John O. Whitaker

The nominations were moved and approved for presentation to the membership.

Program Committee

James Haddock and Douglas Lyng gave the Spring dates at IU-Purdue Fort Wayne as April 22, 23 and for the Fall meeting October 27, 28, 29. Suggestions were requested for speakers and possible topics for the Saturday meeting next year. The IAS Council consisting of the six officers will clear program contributions and policy. By consensus it was felt that we should continue the Saturday meetings and that they may profitably be combined with meetings of other organizations where possible.

Publications Committee

Benjamin Moulton, Chairman, listed September 1983 as the deadline for material to appear in the Centennial volume. We are still looking for an author for a volume possibly titled *Scientists of Indiana.*

Science and Society

Craig E. Nelson, Chairman, was sick and no report was made.

Membership Committee

Alice S. Bennett, Chairman, reported that in October letters were sent to former division chairmen for the years 77 through 80 asking personal contacts with colleagues to recruit new members.

Duvall Jones asked if club memberships could be increased and put on a firmer basis. Furthermore, a concerted effort to bring high school teachers into the Academy should be made, since many do not know that the Academy would be beneficial for them.

Fellows Committee

The report from Richard L. Conklin was presented by John J. Favinger. A motion was made and seconded to accept the list of candidates for presentation at the General Meeting. The motion was approved.

Invitations Committee

President Eberly reported for Donald J. Cook, Chairman, that the 1984 meeting will be at Butler University. Indiana University is not yet confirmed for 1985. Some question was raised about facilities at IU East which has invited the Academy for 1986. The question of a possible registration fee was raised for a future meeting and will be carefully considered in the future.

Emeritus Members Committee

Robert Cooper, Chairman, put in nomination the following people for emeritus membership.

Dr. Howard B. Burkett	1945-1981	37 years
Dr. Marvin Carmack	1953-1982	30 years
John C. Finney	1946-1982	36 years
Maynard K. Hine, D.D.S.	1945-1981	37 years
Dr. Barbara Shalucha	1948-1982	34 years

The nominations were seconded and approved to emeritus status.

It was mentioned that if any of the emeritus members wished to continue to contribute to the Academy that it would be permitted.

REPORTS, SPECIAL APPOINTMENTS

Representative to Indiana Natural Resources Commission

Damian V. Schmelz reported on some of his continuing activities on this Governor's Commission where he has the opportunity to monitor and help shape policies for the wise usage of Indiana's natural resources.

Newsletter Editor

Walter A. Cory, Jr. distributed forms for reporting news for the Newsletter. Mention is again made to the membership that the call for papers will appear in the Newsletter.

A note of appreciation was made to Cory for his editorship of Newsletter Volumes 14 thru 38.

Biological Survey Committee

Theodore J. Crovello reported that the Annual Survey of Literature and Works

in Progress Dealing With Plants, Animals and Environments of Indiana continues. Questionnaires for the 1982 Survey are now available. A questionnaire to establish a Register of Indiana Herbaria is available. The Register will complement the International Index Herbariorum.

Speaker of the Year Committee

Damian Schmelz, Chairman, presented a listing detailing the presentations of our Speaker-of-the-Year at the various colleges and universities in Indiana since 1971.

NEW BUSINESS

President Eberly solicited names to serve on the Committee for the Relation of the Academy to the State. This committee has been inactive for some years but its activities could be particularly important now.

President Eberly also instituted a Special Centennial Program Committee for 1984 to begin planning for the 100th year celebration.

The meeting adjourned at 10:00 p.m.

MINUTES OF THE GENERAL MEETING

October 19, 1982

The meeting was called to order by President Eberly at 1:35 p.m. in the Auditorium of the Center for Continuing Education at the University of Notre Dame.

Robert E. Gordon, Vice President of Advanced Studies, The University of Notre Dame and Fellow of the Indiana Academy of Science presented the welcoming remarks. He pointed out that this was the sixth time the Academy had met on the Notre Dame campus.

John H. Meiser, Secretary, read the minutes. The following nominations were presented to the membership:

President-Elect	Theodore Crovello University of Notre Dame
Secretary	Richard Conklin Hanover College
Research Grants Committee	John Whitaker Indiana State University

After there were no nominations from the floor, nominations were closed and the membership voted to approve the named individuals for their respective positions.

The following individuals were presented for election to the rank of Fellow by John J. Favinger, Representative of the Committee on Fellows.

John A. Bacone, Alice S. Bennett, Robert F. Dale, Jacques W. Delleur, William B. Hopp, Duvall W. Jones, Gayton C. Marks

Certificates were issued to those present.

The minutes were moved for acceptance, seconded and approved.

Ed Miller of the Resolutions Committee presented the following Resolution:

RESOLUTION:

WHEREAS: The Indiana Academy of Science is hightly appreciative to the University of Notre Dame for their invitation to hold its 98th annual meeting on their campus; and

WHEREAS: Students, faculty and administration have cooperated in sharing their facilities with us for this meeting; be it

RESOLVED: That the members of the Academy assembled here express their appreciation to Father Theodore Hesburgh, President of The University of Notre Dame, and to Robert E. Gordon, Vice President of Advanced Studies, for all the courtesies that have been extended to the Academy during this meeting. We wish especially to thank Dr. Theodore J. Crovello and his committee for the arrangements of the entire program and for the comfort and conveniences provided to us. We also express our thanks to the members who organized and are participating in the symposium on "Computers and Value-Oriented Science Education."

This was seconded and approved.

The following are the Division Chairmen and the Chairmen-Elect elected at this meeting.

Anthropology	
Chairman:	Gary D. Ellis
Chairman-Elect:	Donald R. Cochran
Botany	
Chairman:	David Polley
Chairman-Elect:	Phil Pope
Cell Biology	
Chairman:	Zafar Iqbal
Chairman-Elect:	Ralph Jersild
Chemistry	
Chairman:	Eric R. Johnson
Chairman-Elect:	Shannon Lieb
Ecology	
Chairman:	Richard Whitman
Chairman-Elect:	Edwin R. Squiers
Engineering	
Chairman:	David D. Chesak
Chairman-Elect:	Thomas R. Hanley
Entomology	
Chairman:	R. Bruce Cummings
Chairman-Elect:	Paul Robert Grimstad
Environmental Quality	
Chairman:	Robert H. L. Howe
Chairman-Elect:	William Beranek, Jr.
Geology and Geography	
Chairman:	Terry R. West
Chairman-Elect:	Gerald R. Showalter
History of Science	
Chairman:	Gene Kritsky
Chairman-Elect:	Gene Kritsky
Microbiology and Molecular Biology	
Chairman:	Mary Lee Richeson
Chairman-Elect:	J. R. Garcia
Physics and Astronomy	
Chairman:	Malcom Hults
Chairman-Elect:	Vincent A. Di Noto, Jr.
Plant Taxonomy	
Chairman:	James R. Aldrich
Chairman-Elect:	Marion Jackson
Science Education	
Chairman:	Stanley Shimer
Chairman-Elect:	Linda A. Hamrick
Soil and Atmospheric Sciences	
Chairman:	H. Raymond Sinclair, Jr.
Chairman-Elect:	Charles Rhykerd

Zoology

Chairman:	Ronald Richards
Chairman-Elect:	Wynn Cudmore

Fay Kenoyer Daily, necrologist, reported on the following members

G. Bryant Bachman	Robert A. Ragains
Hillis L. Howie	Bernard Henry Schockel
Louis Augustus Krumholz	William John Tinkle
Nathan E. Pearson	Yeram Sarkis Touloukian

President Eberly introduced the speaker, Sherman A. Minton, Jr., M.D., Indiana University Medical Center, Indianapolis. The talk was titled, "Highlights in Herpetology: It's Not All Snakes!"

After a very interesting presentation, the meeting was adjourned at 3:05 p.m.

Respectfully submitted,

John H. Meiser
Secretary

Indiana Academy of Science
FINANCIAL REPORT
1 January - 31 December 1982

I. ACADEMY ACCOUNTS

	Income	Budgeted	Expenditures	Budget
Dues	$ 8,370.00	$ 8,000.00	—	—
Reprints: Vol. 90	2,510.90	3,500.00	$ 2,347.40	$ 3,000.00
Interest	2,026.56	1,500.00	—	—
Miscellaneous	10.00	—	—	—
Secretary	—	—	901.40	600.00
Treasurer	—	—	751.75	600.00
General Office	—	—	0.00	250.00
Officer Honorarium	—	—	150.00	150.00
Membership Committee	—	—	34.33	200.00
President's Contingency Fund	—	—	69.00	225.00
Newsletter	—	—	750.00	750.00
Speaker of the Year Honorarium	—	—	700.00	700.00
Program Committee	—	—	3,164.49	1,900.00
AAAS Representative	—	—	0.00	100.00
Public Relations	—	—	150.00	150.00
Editor's Expense	—	—	700.00	700.00
Youth Activities	—	—	0.00	400.00
Biological Survey Committee	—	—	0.00	2,250.00
Section Chairmen's Expenses	—	—	0.00	100.00
CPA Fees for Tax Returns	—	—	775.00	800.00
Miscellaneous (Research Committee)	—	—	100.00	100.00
Transfers to Administered Accounts	—	—	2,700.00	2,700.00
Junior Academy ($400.00)				
Science Talent Search ($500.00)				
Library Binding ($1,500.00)				
Proceedings: Mailing ($300.00)				
TOTAL	$12,917.46	$13,000.00	$13,293.37	$15,675.00

II. ADMINISTERED ACCOUNTS

	January 1 Balance	1982 Transfers & Income*	1982 Expenditures	Dec. 31 Balance
Junior Academy	$ 1,230.66	$ 400.00 (T1)	$ 418.90	$ 1,211.76
Science Talent Search	295.43	500.00 (T1)	2,774.25	619.18
		2,598.00 (I)		
Science & Society	1,447.89	0.00	0.00	1,447.89
Research Grants	(-6,331.51)	18,963.00 (T2)	19,803.00	-6,271.51
		900.00 (I)		
Natural Areas	0.00	0.00	0.00	0.00
J.S. Wright Library Fund	134.28	0.00	0.00	134.28
Lilly III Library Fund	2,632.76	0.00	0.00	2,632.76
Lilly V Library Fund	4,500.20	0.00	0.00	4,500.20
Library Binding	1,202.55	1,500.00 (T1)	1,201.90	1,500.65
Proceedings: Publication	1,561.89	5,522.25 (T2)	5,522.25**	1,589.89
		28.00 (I)		
Proceedings: Mailing	822.75	300.00 (T1)	365.88	756.87
Monographs	2,346.20	13,009.08 (T2)	13,438.92	2,964.91
		1,048.55 (I)		
Dept. of Interior Grant 14-16-0003-79-133	(-595.36)	2,698.25 (I)	2,102.89	0.00
TOTAL	$ 9,247.74	$47,467.13	$45,627.99	$11,086.88

*I: Income from external sources. T1: Transfer from Academy Accounts.
T2: Transfer from Academy Trust Funds.

** The State of Indiana paid an additional $8,000 toward printing of the *Proceedings*.

III. SUMMARY

	Academy Accounts	Administered Accounts	Totals
Balance: 1 January 1982	$ 9,041.08	$ 9,247.74	$18,288.82
1982 Income & Transfers	12,917.46	47,467.13	60,384.59
1982 Expenditures	13,293.37	45,627.99	58,921.36
Balance: 31 December 1982	8,665.17	11,086.88	19,752.05

IV. BANK BALANCES

Checking Account	
Farmers and Merchants Bank, Rensselaer, IN	$ 1,489.82
Savings Accounts (Principal as of 31 December)	
Farmers and Merchants Bank, Rensselaer, IN—C.D.	5,522.12
Farmers and Merchants Bank, Rensselaer, IN—Passbook	379.64
State Bank of Rensselaer, IN—C.D.	12,360.47
TOTAL	$19,752.05

V. SUMMARY OF TRUST FUNDS

A. Foundation Account (00430-00-0)

1. Income Account			
Income cash balance (1/1/82)		$ 0.00	
Dividends and interest for 1982		2,463.07	
Disbursements for 1982			
Research grants (Received in 1983)	$ 300.00		
Transfer to Principal cash	2,163.07		
	$ 2,463.07	-2,463.07	
Income cash balance (12/31/82)		$ 0.00	0.00
2. Principal Account			
Principal cash balance (1/1/82)		$ 1,028.60	
Total receipts for 1982		19,000.00	
Transfer from Income Account		2,163.07	
Total disbursements for 1982			
Investments purchased	$21,900.00	-21,900.00	
Principal cash balance (12/31/82)		$ 291.67	291.67
Market value of investments (12/31/82)			$35,105.42
Total value of account (12/31/82)			$35,397.09*

*Carrying value of investments (12/31/82) is $32,081.50

B. John S. Wright Fund (00430-01-9)

1. Income Account			
Income cash balance (1/1/82)		$ 5,720.36	
Dividends and interest for 1982		36,388.99	
Disbursements for 1982			
Commission and fees	$ 3,527.00		
Miscellaneous expenses	37,069.85		
	$40,596.85	-40,596.85	
Income cash balance (12/31/82)		$ 1,512.50	$ 1,512.50
2. Principal Account			
Principal cash balance (1/1/82)		$ 112.15	
Total receipts for 1982		42,076.93	
Total disbursements for 1982		22,179.31	
Principal cash balance (12/31/82)		$20,009.77	$ 20,009.77
Market value of investments (12/31/82)			662,945.47
Total value of account (12/31/82)			$684,467.74*

*Carrying value of investments (12/31/82) is $296,751.79

C. J.S. Wright Invested Income Account (00430-02-8)

1. Income Account		
Income cash balance (1/1/82)	$ 2,268.75	
Total interest for 1982	13,750.22	
Disbursements for 1982	0.00	
Transfer from Principal Account	684.97	
Transfer to Principal Account	10,778.94	
Income cash balance (12/31/82)	$ 5,925.00	$ 5,925.00
2. Principal Account		
Principal cash balance (1/1/82)	$ 1,102.10	
Total receipts for 1982	161,869.85	
Disbursements for 1982		
Investments purchased	100,681.25	
Distributions (Grants & *Proceedings*)	37,494.33	
Transfer from Income Account	10,778.94	
Transfer to Income Account	684.97	
Principal cash balance (12/31/82)	34,890.34	$ 34,890.34
Market value of investments (12/31/82)		59,053.00
Total value of account		$ 99,868.34

VI. NOTES

Membership dues: The Treasurer's records show that the Academy has 1069 paid memberships for 1982: 33 sustaining, 2 sustaining family, 535 senior, 28 senior family, 251 regular, 14 regular family, 85 student, 102 emeritus, 2 honorary, 3 life and 14 club memberships.

7 members deceased (included in totals above).
93 members on file from 1981, but not paid for 1982.
114 new members for 1982 (included in totals above).
4 previous members reinstated in 1982 (included in totals above).
7 persons resigned.
65 individuals and one club dropped for nonpayment of 1981 dues.

Dues Structure for 1982:

$ 2.00	for student memberships
5.00	for memberships and club memberships
10.00	for senior memberships
25.00	for sustaining memberships
2.00	additional for family memberships
300.00	for life memberships
150.00— 500.00	corporate memberships
50.00— 100.00	institutional memberships
1.00	initiation/reinstatement fee ($2.00 for family membership)

Reprints: All authors of papers in Volume 90 of the *Proceedings* have paid for the reprints which they ordered. Cost of reprints to the Academy was $2,347.40. Authors paid the Academy $2,510.90 for reprints.

Publications: The monograph on *Ectoparasites of Mammals of Indiana* by John Whitaker was printed and released by the Academy in October, 1982. Sales of reprints, monographs and *Proceedings* in 1982 totaled $3,587.45.

Research Grants: Funds totaling $19,803.00 have been awarded to: A.M. Berquist (Notre Dame), R.E. Brown (Huntington C.), J.B. Cave (Ball State U.), T.A. Fogle (St. Mary's C.), R.W. Greene (Notre Dame), J.K. Greenlee (Notre Dame), A.J.C.L. Hogarth (DePauw U.), A.L. Hughes (Indiana U.), K. Kamo (Indiana U.), D.T. Krohne (Wabash), J.R. Litton (St. Mary's C.), M.J. Mandracchia (Notre Dame), C.E. Mays (DePauw U.), N.J. McCreary (Notre Dame), E.D. Mould (St. Mary's C.), V.L. Riemenschneider (IUSB), R. Schwarzwalder (Indiana U.), C.A. Stewart (Indiana U.), B.K. Swartz (Ball State U.), J.S. Wasser (Indiana U.), C. Carter (Purdue U.), S.W. Dhawale (Indiana Univ. East), Vas Dev, (Notre Dame), C.T. Huang (Purdue U.), J.C. Latino (Notre Dame), P.C. MacMillen (Hanover CO.), J.P. Rybarczyk (Ball State U.), D.M. Sever (St. Mary's C.), M. Vaughn (Indiana State U.), H.P. Weeks, Jr. (Purdue U.), S.C. Yoder (Goshen C.), Alan Engel and Nevin Longenecker (John Adams High School), James A. Barsic and Dean Christakis (Marquette High School), Susan Rucinski and M. Kobe (Gavit High School), Tamara M. Hughes and Mrs. Nell Walthers (Plainfield High School), Valori Miller, Jeff Medwetz, Scott Bearby, and M. Kobe (Gavit High School), Jim Szalony and M. Kobe (Gavit High School), Laura Warot and M. Kobe (Gavit Senior High School), Steve

Longenecker and John Koellner (Riley High School), and Kathleen Ramos and M. Kobe (Gavit High School).

Grants Received: The final installment ($2,698.25) of the U.S. Department of Interior Grant 14-16-0003-79-133 for biological survey was received and disbursed.

Kappa Kappa Kappa Sorority made $2,500 available for awards and expenses for the Science Talent Search for Indiana high school students.

The American Association for the Advancement of Science granted $900.00 to be used by high school students for research.

State Audit: The State Board of Accounts has examined the records of the Academy for the period July 1, 1977 to June 30, 1981, and certified that the records are satisfactory.

VII. BUDGET FOR 1983

The following budget was approved by the Budget Committee in the meeting of 4 December 1982.

Academy Accounts	
Anticipated Income	
Dues, Initiation and Reinstatement Fees	$ 8,000.00
Interest on Savings	1,750.00
Reprint Charges to Authors: Vol. 91	3,250.00
	$13,000.00
Budgeted Expenditures	
Secretary	$ 600.00
Treasurer	600.00
General Office	250.00
Officer Honoraria	150.00
Membership Committee	400.00
President's Contingency Fund	225.00
Newsletter	800.00
Speaker of the Year	700.00
Program Committee	2,300.00
Editor's Expenses	700.00
Youth Activities Committee	400.00
Biological Survey Committee	1,000.00
Centennial Committee	200.00
AAAS representative	250.00
Division Chairmen's Expenses	50.00
Public Relations/Research Grants	250.00
CPA Fees for Tax Returns	500.00
Miscellaneous	100.00
Reprint Charges to Academy	2,850.00
Transfers to Administrated Accounts	2,450.00
Science Talent Search ($500.00)	
Library Binding ($1,500.00)	
Proceedings: Mailing ($450.00)	
	$14,775.00
Endowment Funds	
Anticipated Income	
IAS Foundation	$ 300.00
J. S. Wright Invested Income Account	35,000.00
Invested Income carryover	88,500.00
	$123,800.00
Budgeted Expenditures	
Bank Fee	$ 3,500.00
Research Grants for Senior Academy Members	21,000.00
Research Fellowships for Secondary School Teachers (who are Academy Members)	3,000.00
Publications	
Proceedings—Volume 91 ($13,218—8,000)	5,218.00*
	$ 32,718.00

* Anticipated funds of $8,000 from State of Indiana.
Expenses for monographs in preparation will come from the Endowment Funds.

Restricted Accounts	
Anticipated Income	
AAAS Funds for High School Research Grants	$ 900.00
Tri-Kappa Funds for Science Talent Search	2,000.00
Sale of Publications	
Proceedings	50.00
Monographs	750.00
	$ 3,700.00
Anticipated Expenditures	
Research Grants awarded to Junior Academy members	$ 900.00
Science Talent Search	2,000.00
Publications	
Proceedings (part)	50.00
Monographs (part)	250.00
	$ 3,200.00

Respectfully submitted,

Duvall A. Jones, Treasurer

We, the undersigned, have audited the Treasurer's records for the Indiana Academy of Science for the year 1982 and have found them to be accurate and in order.

Andrew G. Mehall

_____, 1983

John Ricketts

_____, 1983

INDIANA JUNIOR ACADEMY OF SCIENCE

OFFICERS

President ..... Jody Sibbitt, Paoli Jr.-Sr. High School
President-Elect ..... Rick Karr, Highland Sr. High School
Secretary ..... Michelle Trinkle, Paoli Jr.-Sr. High School

JUNIOR ACADEMY COUNCIL

Jr. Scientist, Sr. Division .....Thomas Anderson, Paoli Jr.-Sr. H.S.
Chairpersons
.....Michael Kobe, Gavit High School, Hammond
Jr. Scientist, Jr. Division ..... William Fariss, Gavit Middle School
Chairpersons
.....Laural Krool, Gavit Middle School
Senior Papers, Chairperson .....Jane Tucker, St. John the Baptist Fort Wayne
Junior Papers, Chairperson .....Virginia Rhodes, East Noble High School Kendallville
Polemics, Chairperson .....Cheryl Mason, Highland High School Highland
Science Fiction, Chairperson .....Leota Skirvin, Brown County High School Nashville
Poster Papers .....Joanne Richardson, Pat Fleming, Eileen Ridley
Jr. and Sr. Divisions University of Notre Dame
Mousetrap Cars .....Steve Makurat, Brown County High School
Steve Brown, University of Notre Dame
Paper Planes .....Steve Kraemar, Richard Bush
University of Notre Dame

PROGRAM

(Notre Dame Liason: Reverend James J. McGrath, Professor of Biology)

Thursday, October 28

7:30-11:00 PM	Check in at Stepan Center

Friday, October 29

8:30-11:00 AM	Registration, Stepan Center
9:00- 9:30 AM	Welcome by Dr. Daniel H. Winicur, Assistant Dean of the College of Science
	Dr. William Eberly, President of the Indiana Senior Academy of Science, congratulates the Jr. Academy on its 50th Anniversary
	Introduction of Candidates and Announcements
9:40-11:30 AM	Poster Paper Session, Stepan Center

9:40-10:30 AM	Paper Plans and Mousetrap Car Contests, Stepan Center
9:40-	Junior Scientist Interviews, Stepan Center
10:00-11:30 AM	Presentation of Papers, Jr. Division Room 355, Galvin Life Sciences
	Presentation of Papers, Sr. Division Room 357, Galvin Life Sciences
10:15-10:55 AM	Group "A" Polemic, Stepan Center
11:00-11:40 AM	Group "B" Polemic, Stepan Center
12:00- 1:00 PM	LUNCH
1:30PM	Presentation of Awards Election of Officers
2:45 PM	Adjournment

ELECTED STATE OFFICERS 1983-1984

President-Elect ..... Petra Fuerhaupter, Highland Sr. High School
Secretary ..... Joseph V. Pawlowski, Highland Sr. H.S.

CANDIDATES FOR STATE OFFICE 1983-1984

President-Elect ..... Petra Fuerhaupter, Highland Sr. High School
Secretary ..... Joseph V. Pawlowski, Highland Sr. H.S.

*SENIOR DIVISION PAPERS
*JUNIOR DIVISION PAPERS
*NOMINATIONS FOR INDIANA JUNIOR SCIENTIST—SR. DIVISION
*NOMINATIONS FOR INDIANA JUNIOR SCIENTIST—JR. DIVISION
*SCIENCE FICTION STORIES—SR. DIVISION
*SCIENCE FICTION STORIES—JR. DIVISION

*The above information is on the enclosed pages.

NOMINATIONS FOR INDIANA JUNIOR SCIENTIST

SENIOR DIVISION

CENTRAL NOBLE HIGH SCHOOL—Jennifer Farr
PAOLI JR.-SR. HIGH SCHOOL—Jody Lynn Sibbitt
GAVIT JR.-SR. HIGH SCHOOL—Marc Hockman, Jennifer Kucharski
HIGHLAND HIGH SCHOOL—Anne Tseng
MARQUETTE HIGH SCHOOL—Jim Barsic, Ali Raheem
INDIAN CREEK HIGH SCHOOL—Michael Fell

NOMINATIONS FOR INDIANA JUNIOR SCIENTIST

JUNIOR DIVISION

THE CANTERBURY SCHOOL—Christopher Moses
GAVIT JR.-SR. HIGH SCHOOL—James Warner Huggler, Jr.
MARQUETTE HIGH SCHOOL—James Jensen, Mark Owens

SENIOR DIVISION PAPERS

MARQUETTE HIGH SCHOOL

Jim Barsic—"Scanning Electron Microscope Studies of the Effectiveness of HIP Processing and Single Crystal Components in Reducing Stress Fractures and Cleaving on Industrial and Military Turbine Components."

Ali Raheem—"Research into Hyperglycemia: A Comparison of R-Regular Iletin Insulin and an Author-Extracted Mouse Insulin on the Blood and Urine Glucose Levels of the C57BL/BL?KsJ-dbm Diabetic Strain of Mouse."

CENTRAL NOBLE HIGH SCHOOL

Jennifer Farr—"The Effect of Trazco B*401 on the Decomposition of Herbicidal Residues Under Specific Environmental Conditions"

PAOLI JR.-SR. HIGH SCHOOL

Heidi White—"The Direct Effects of Glucocorticoids *In Vitro* on Populations of Lymphocytes"

Jody Lynn Sibbitt—"Possible Contributory Factors to Toxic Shock Syndrome"

EAST NOBLE HIGH SCHOOL

Nanette Latchow—"The Effects of Volcanic Ash versus Silica on the Mammalian Respiratory System"

GAVIT JR-SR. HIGH SCHOOL

Marc Hochman—"The Isolation and Revival of Clostridium"

Jennifer Kucharski—"The Effects of Food Stuffs Prepared in a Microwave Oven on Bacteria Commonly Found in the Human Digestive System."

HIGHLAND HIGH SCHOOL

Ernst Tseng—"The Role of Testosterone in Regulating Rat Acid Phosphatase Activities and Specific Gene Expression"

Kelly Ward—"Identification of the Site of Esterase 6-Synthesis in Drosophila melanogaster"

JUNIOR DIVISION PAPERS

MARQUETTE HIGH SCHOOL

Mark Owens—"Holographic Interferometry: Measurement of Pressure-mediated Micro-movement in Various Metals"

James Jensen—"Parabolization of an 8-inch, F-5, Primary Mirror to be Integrated in a Cassegrain-Newtonian Reflecting Telescope."

GAVIT JR.-SR. HIGH SCHOOL

James W. Huggler, Jr.—"The Burning Question"

Paul Chojenski—"Catching the Sun's Energy"

EAST NOBLE HIGH SCHOOL

Laura Emerson—"Burning Wood, an Energy Alternative"

THE CANTERBURY SCHOOL

Debbie Parrott—"Interaction of Ions: Do All Metals and Non-metals Interact?"

Christopher Moses—"In Quest of the Photon: Laser-induced Holographic Images"

SAINT JOHN THE BAPTIST

Paul Sinnes—"Telephone Communication by the Use of Laser Light"

SCIENCE FICTION STORIES

SENIOR DIVISION

HIGHLAND HIGH SCHOOL

Todd Gyure—"The Selection Line"

Kelly Ward—"The Drosophila Disaster"

GAVIT JR.-SR. HIGH SCHOOL

Jennifer Kucharski—"The Cold Sleep"

MARQUETTE HIGH SCHOOL

Tom Totten—"The Map"

Jim Barsic—"New World Man"

Patrick Bobillo—"The Silver Bear"

JUNIOR DIVISION

HIGHLAND HIGH SCHOOL

Charles E. Wolven—"The Encounter"

GAVIT JR.-SR. HIGH SCHOOL

Marc Vockell—"Chips"

SAINT JOHN THE BAPTIST

Mary Brita—"The Cloning of Mary Brita"

Janet Libbing—"The Demanding Muscles"

THE CANTERBURY SCHOOL

Stanton W. Clark—"Molecular Manipulation"

Renata Bauman—"The Great Explosion"

MARQUETTE HIGH SCHOOL

Mark Owens—"Light Traveler"

Eric Bonfield—"The Abogenitive Crisis"

Lisa Totten—"Speculations"

James Jensen—"A Matter of the Mind"

SENIOR DIVISION

OUTSTANDING SCIENTIST ..... Jody Lynn Sibbitt, Paoli Sr. High School

PAPER PRESENTERS .....
1st— Jennifer Farr, Central Noble
2nd— Jody Sibbitt, Paoli Jr.-Sr. H.S.
3rd— Kelly Ward, Highland Sr. H.S.

SCIENCE FICTION STORIES .....
1st— Todd Gyure, Highland
2nd— Jim Barsic, Marquette
3rd— Kelly Ward, Highland

POLEMICS .....
1st— Marquette High School
2nd— Highland High School
3rd— Marquette High School

PAPER PLANE CONTEST .....
1st— Marquette High School
2nd— Gavit High School
3rd— Gavit High School

MOUSETRAP CAR CONTEST .....
1st— Highland High School
2nd— Marquette High School
3rd— Highland High School

POSTER PAPERS .....
1st— Chris Brown, Highland H.S.
2d— Petra Fuerhaupter, Highland H.S.
3rd— Ali Raheem, Marquette H.S.

JUNIOR DIVISION

OUTSTANDING SCIENTIST ..... Mark Owens, Marquette

PAPER PRESENTERS .....
1st— James Jensen, Marquette
2nd— James Huggler, Jr.
3rd— Laura Emerson, East Noble

SCIENCE FICTION STORIES .....
1st— Mary Brita, St. John The Baptist
2nd— Renata Bauman, The Canterbury School
3rd— Stanton Clark, The Canterbury School

POLEMICS .....
1st Marquette
2nd— Highland
3rd— Gavit

PAPER PLANE CONTEST .....
1st— Sean Evisten, Canterbury
2nd— Steve Rogers, Gavit
3rd— Allan Spott, Highland

MOUSETRAP CAR CONTEST .....
1st— Allan Spott, Highland
2nd— Eric Bonfield, Marquette
3rd— Erin Dellinger, The Canterbury School

POSTER PAPERS .....
1st— Heather Hamrick, The Canterbury School
2nd— Stan Clark, The Canterbury School
3rd— Debbie Parrott, The Canterbury School

Necrology

Fay Kenoyer Daily, Butler University

G(USTAVE) BRYANT BACHMAN

Kansas City, Missouri
August 18, 1905

Lafayette, Indiana
April 14, 1982

Dr. Gustave Bryant Bachman was an Emeritus Professor of Chemistry at Purdue University who died April 14, 1982, at Lafayette, Indiana. He had been in ill health several months.

Born at Kansas City, Missouri, August 18, 1905, he attended Kemper Military School in Boonville, Missouri. Later, he lived in New Mexico and then Colorado where he received an A.B. degree, Magna Cum Laude from Colorado University in 1926. A Ph. D. degree from Yale followed in 1930 as well as some post graduate work at Munich Germany from 1929 to 1930.

Dr. Bachman's teaching career began at Ohio State University in 1930 as an instructor and continued from 1933 to 1936 as assistant professor. In 1936, he joined Eastman Kodak in New York, but returned to teaching in 1939 as an Associate Professor at Purdue University in Lafayette, Indiana. He became a Professor of Chemistry in 1941. When he retired in 1971, he remained active in campus life and retained an office there. During his teaching career, he trained about 75 students for the Ph. D. degree and 75 for an M.S. degree. He initiated courses in Theories of Organic Chemistry, Natural Products and Heterocyclic Chemistry for graduate students. He published the textbook, *Organic Chemistry*, in 1949.

Dr. Bachman also served in various university affairs chairing the Committee for Reorganization of the Faculty, Committee Structure and Committee for Writing a Constitution for the School of Science. He was a member of the Convocation Committee and University Senate Faculty Affairs Committee. He was instrumental in founding a Phi Beta Kappa Chapter at Purdue. He also served as a consultant for several chemical companies including Commercial Solvents, Eli Lilly and Company, Diamond Alkali Company, Continental Carbon Company, General Tire and Rubber Company and others.

Dr. G. Bryant Bachman was internationally known for his research in the organic chemistry field with special interest in aliphatic nitro compounds and the mechanism of combustion. Included, too, were plastics, rubbers, antimalarials and other drugs, nitration and synthetic organic chemistry. He published over 100 papers and obtained about 35 patents.

Outside his academic life, Dr. Bachman was interested in music becoming the founder and first President of the Lafayette Symphony. He was a member of the Rotary Club and gave lectures to them on Astronomy in which he also was interested. He liked sailing and was associated with the Indiana Power Squadron in Indianapolis. This interest gave rise to his writing a witty book after retirement. It was called *Definitions: Nautical and Naughtical.*

Dr. Bachman joined the Indiana Academy of Science in 1939 when he came to teach at Purdue University in Indiana, and was honored as Fellow in 1952. He was an Emeritus Member at death. He gave several papers at meetings: one

on a college undergraduate course in organic chemistry, another (co-author) on hydrolysis rates of unsaturated esters in alkaline emulsion systems of the GR-S type, and also on (co-author) synthetic resins and base exchange phenomena. He was Chairman of the Division of Chemistry of the Academy in 1967 and served on the Executive Committee. He was a member of the University Club, American Chemical Society (local), Sigma Xi, Phi Beta Kappa and was in elected offices of the St. John's Episcopal Church.

Besides the *American Men and Women of Science*, we are in debt to colleagues of Dr. Bachman at Purdue, Harry Morrison (chairman), M.G. Mellon and Richard Sneen for sharing biographical information about Dr. Bachman. We not only received the memorial resolution from Dr. Morrison, but a copy of the fine article in the *Journal and Courier*, Lafayette, Indiana, April 15, 1982, which were very helpful in writing this report.

HILLIS LANGHORNE HOWIE

Indianapolis, Indiana
January 31, 1903

Bloomington, Indiana
June 2, 1982

Mr. Hillis Howie was a school administrator, teacher and director of expeditions to the southwest United States who died at Bloomington, Indiana, June 2, 1982.

Born in Indianapolis, Indiana, January 31, 1903, most of his early education was obtained there, too, and he graduated from Shortridge High School. He studied at Wesleyan University at Middletown, Connecticut, for two years and then obtained a B.A. degree from Butler University in 1925. He worked on a Master's degree many years later obtaining an M.A. at Yale University in 1949.

Before his years as administrator, Mr. Howie was a teacher in woodcrafts and nature study for eight years at Culver Military School beginning while he was a high school student. From 1926 to 1928, he was a teacher at Orchard School in Indianapolis, Indiana. From 1928 to 1929, he taught at Dalton School in New York City. From 1930 to 1933, he was back teaching at Orchard School. From 1933 to 1938, he served as Director of Orchard School where he was much loved and admired. He was a good teacher and had a great influence on young people. He resigned in 1938 to study at Yale University. He was Chairman of the School Service Department of the Peabody Museum of Natural History at Yale from 1942 to 1946. In 1946, he was appointed Director of the Community School in St. Louis, Missouri, where he stayed until 1959. He was Director of Poughkeepsie Day School in New York from 1963 to 1965.

Mr. Howie may be as well known for the Prarie Trek Expeditions which he organized in 1926 in order to take boys on camping trips to the southwestern Unites States of America. We learn from *Keep an Eye on that Mummy*, a history of the Children's Museum of Indianapolis, by Nancy Kriplen that the trips were fashioned after camping trips to the west taken by boys Mr. Howie had known while a counselor at Culver's summer Woodcraft School. Most of the boys were members of a Boy Scout Troop which he had led and there were nine that first year. Mr. Stewart Springer (now an internationally known authority on sharks and a former Indiana Academy of Science member) was drafted as an assistant. He had left the Children's Museum a few months earlier where he had been the Museum Curator. His interest in collecting and preserving natural history specimens led to instructing the boys in the rudiments of good zoological techniques. Mr. Howie saw how the trips could be more than sightseeing events and provide a sound scientific education in various scientific fields and journalism (chosen by some who were responsible for sending reports back to the *Indianapolis Star* newspaper). The *Indianapolis Star* published a good article May 16, 1933, about Mr. Howie and the treks. Also, one appeared in the May 7, 1960, *Indianapolis News*.

In 1928, Mr. Howie left Orchard School to go to New York City to teach, but the next summer, he took the Prarie Trek expedition to England as a delegation from Indianapolis to an International Boy Scout Jamboree. He was "disillusioned" by the affair and conducted the treks after that without Boy Scout emphasis.

Back in Indianapolis at Orchard School in 1930, Mr. Howie offered to make the trek a Children's Museum expedition to bring back archeological and natural history exhibits for the museum. The trek atracted much attention. He had also made the first seahorse emblem for the Museum while a teacher of shop at Orchard

School. It was designed by Kurt Vonnegut. Mr. Howie had been on the Museum Board for a year before going to New York and had collected small mammals for the Washington National Zoo and American Museum of Natural History.

In 1934, the Turquoise Trail headed west. This was fashioned after Mr. Howie's expeditions and was for girls. It was headed by Gordon Thompson, a former assistant of Mr. Howie.

Mr. Howie bought 500 acres called Cottonwood Gulch near Thoreau, New Mexico in 1934. He used the ranch as headquarters for the expeditions of which he was Director from 1926 to 1970 when he retired and was Chairman of the Board of Trustees until 1977. Mr. Kurt Vonnegut designed the cabins built there in exchange for tuition for his son, Kurt Vonnegut, Jr., to attend the 1937 trek. The camp and expeditions were incorporated as a non-profit educational foundation, the Cottonwood Gulch Foundation.

Mr. Howie was back on the Children's Museum Board in 1931 and was first vice president from 1933 to 1937. The Museum was a point of departure for the treks until 1942 and World War II which caused a temporary halt in the activity.

Mr. Howie's hobbies were numerous such as woodcrafts, birds and antique tools. He was author of several articles on out-of-door education.

Mr. Howie joined the Indiana Academy of Science in 1935 and was an Emeritus Member at death. He was also a member of Psi Upsilon Fraternity, Indiana Historical Society and Unitarian Universalist Church of Bloomington, Indiana.

Mr. Howie died in Bloomington, Indiana, June 2, 1982. His interest in young people is the thread woven through all the accounts of his life upon which this report is based. Besides the Children's Museum history cited above, material was kindly sent us by Mrs. Howie and son, John.

LOUIS AUGUSTUS KRUMHOLZ

Harrington, Washington
December 1, 1909

Louisville, Kentucky
January 23, 1981

Dr. Louis A. Krumholz was a retired educator, research scientist in biology and administrator who died January 23, 1981, in his Louisville home. He was the subject of numerous articles in the *Courier Journal* of Louisville, many of which were provided for us by Mr. Dwayne Cox, archivist, at the library of the University of Louisville. These and other records from the archives provided much of the information here.

Born at Harrington, Washington, December 1, 1909. When a few years old, Dr. Krumholz moved to Minnesota where he spent his boyhood. He lived in Fairmount, an agricultural community, near a chain of lakes where he became fascinated with the aquatic life he saw. A B.S. degree was obtained from the College of St. Thomas in St. Paul, Minnesota in 1932. At that time, he wanted to be a bone surgeon and entered medical school at Marquette University. After two years, he could not continue because of the deepening depression and his lack of funds. He began teaching general science and English at a small consolidated school in Minnesota and decided to study German. However, he returned to biology after two years with a renewed interest developed while working with Dr. Josephine Tilden on her algal studies. He obtained an M.S. degree from the University of Illinois in 1938 and a Ph. D. in zoology at the University of Michigan in 1945.

While at Illinois, he was assistant zoologist with the State Natural History Survey from 1937 to 1940. He was Aquatic Biologist with the Michigan Institute for Fisheries Research at Ann Arbor from 1940 to 1945. He came to Indiana in 1945 to be an instructor in Zoology at Indiana University and was Research Associate of the Indiana Lake and Stream Survey until 1950. He then went to Oak Ridge, Tennessee, with the Tennessee Valley Authority as Aquatic Biologist from 1950 to 1954. He was Technical Reports Analyst for Oak Ridge TVA in 1954. He then moved to the Bahamas where he was Resident Biologist in Charge of the Lerner Marine Laboratory on Bimini from 1954 to 1957. In 1957, he went back to teaching at the University of Louisville in the Department of Biology. He became Director of the Water Resources Laboratory in 1967 where he served until 1975. From 1975 until he retired in 1979, he was Assistant Vice President for Academic Affairs.

Dr. Krumholz was a very active and productive teacher and was also active in academic affairs. He was an internationally known aquatic ecologist and radiation biologist. His research included population studies of fishes, embryology, comparative anatomy and fecundity of fishes, the effect of radioactive wastes on aquatic fauna and the accumulation and selective concentration of radioactive materials in aquatic vertebrates. He was author or co-author of over 100 scientific articles and had around 25 graduate students whose scientific research was directed by him. His work at TVA attracted great interest and was reported in the *New York Times*, July 1, 1956, because he concluded that radioactive waste from the atomic energy plant was cumulative in fish shortening their life span and speed of growth. He received many honors. Perhaps the most important one was bestowed by the *Wildlife Society* for his outstanding publication in wildlife ecology and management 1955 to 1956 and Honorary Membership for outstanding achievement and service. He also received the Kentucky Academy of Science Distinguished Scientist Award in 1976. He was invited to participate in many national and international symposia as the one on radiation ecology in Stockholm in 1966 and the

British Ecological Society in Northern Ireland in 1975. His activities were often of public interest and were reported in newspapers as mentioned above and the *Louisville Times* reported on his work. He attracted international interest in 1967 when he was a consultant in a study of the effect on wildlife and food supply that the use of underground nuclear power might have if used for blasting for a new Panama Canal. He was an editor for the *Journal of Wildlife, Ecology, Journal of Fisheries Research Board of Canada, Copeia,* and *Wildlife Monographs.*

Dr. Krumholz joined the Indiana Academy of Science in 1969 and presented a paper that year on Mussels of the Wabash and White Rivers of Indiana. He was also a member of the American Society of Ichthyologists and Herpetologists, American Fisheries Society, American Society of Limnology and Oceanography, American Association for the Advancement of Science, American Institute of Biological Science, American Society of Zoologists, American Association of University Professors, Ecological Society of America, British Ecological Society, Wildlife Society, American Institute for Fisheries Research Biology, Gulf and Caribbean Institute, Kentucky Academy of Science and International Society of Limnology.

A list of 75 publications by Dr. Krumholz was prepared while he was Director of Water Resources Laboratory at the University of Louisville. It ended with a 1972 paper and was provided for our files by Mr. Cox.

NATHAN E(VERETT) PEARSON

Amboy, Indiana
December 12, 1895

Hamilton, Indiana
September 25, 1982

Dr. Nathan E. Pearson was a native of Indiana and former head of the Zoology Department of Butler University, Indianapolis, Indiana.

Born at Amboy in 1895, Dr. Pearson received much of his education in the state. He attended Jonesboro, Indiana, High School from which he graduated in 1915. He earned an A.B. degree in 1921, an A.M. in 1923 and a Ph. D. in 1926 at Indiana University, Bloomington, Indiana. He also studied at Indiana State Normal College at Terre Haute, Indiana, for 24 weeks in the summers of 1915 and 1916. The summers of 1919 and 1920, he was at the Indiana University Biological Station. He was a student at Lake Forest College, Lake Forest, Illinois, from 1917 to 1918. He served as a private in the U.S. Army during World War I from 1918 to 1919.

Dr. Pearson's teaching career began in rural schools of Indiana in 1915. At that time, a high school graduate could take 12 weeks normal training in the summer and teach in the fall. He was at Woods Hole Biological Station in Massachusetts the summer of 1925 and was teaching at Columbia University from 1925 to 1926. He was an assistant in Zoology at Indiana University from 1921 to 1925. He served as Ichthyologist of the Mulford Biological Exploration in Peru, Bolivia, and the Amazon Basin in Brazil from 1921 to 1922, and was a collector for Indiana University on an expedition to Peru, South America in 1923. He collected specimens for University of Michigan on both of the above expeditions, too. He became quite interested in the taxonomy and distribution of South American fresh-water fishes.

Dr. Pearson began teaching at Butler University in 1926 in the Zoology Department. The summers of 1926 and 1927, he was Professor of Biology and Nature Study at Winona Normal School. He became Head of the Department of Zoology at Butler University in 1938 in which capacity he continued until retirement in 1964. He was well-liked and a very effective teacher. His patience, even temper and genuine interest in young people helped him establish a good rapport with students. His lectures were well-organized and the material was current. In 1963, he received the Butler Medal, the highest award given by the Butler Alumni Association, for distinguished service. He taught and was adviser to many pre-medical students and more than 250 of them earned a doctor of medicine degree. Many other students who did not go into the medical profession have distinguished themselves in other fields after preparation under his guidance. He also wrote many articles on fishes. He was listed in *Who's Who of America* and *Who's Who in the Midwest, American Men and Women of Science, Indiana Authors and their Books 1917-1966* by D.E. Thompson, *Indiana Scientists* by S.S. Visher, articles in the *Butler Alumnus* and *Indianapolis Star* and *Indianapolis News.*

Dr. Pearson joined the Indiana Academy of Science in 1922 and became a Fellow in 1931. He was an Emeritus Member at death. He gave several papers at meetings on the cytology of a group of katydids, a disease in fishes and the reproductive cycle of an infusorian. He was very active for about thirty years serving on the Publication of the Proceedings, Program (twice), Auditing, Biological Survey, Membership, Fellows, and Executive Committees. He was also on a special committee on undergraduate research in biology and was Vice-president of the Indiana Academy of Science in 1946. In addition, he was a member of Phi Kappa

Phi, Sigma Xi, the American Society of Zoologists, American Association of University Professors, American Association for the Advancement of Science and Fellow. The Blatchley Nature Study Club made him an Honorary Member.

During Dr. Pearson's later years, he spent the summers on a farm near Hamilton, Indiana, the home of Patty Douglas, his daughter, who provided much of the information for this article. There he followed one of his hobbies, gardening, to the envy of the neighborhood. He had many other interests, too, as repairing old clocks, geneaology, woodworking, and many others following his natural curiosity down many avenues with enthusiasm. Another daughter, Barbara Scott, survives him.

It was while visiting the farm that Dr. Pearson had a heart attack and died suddenly September 25, 1982.

ROBERT A. RAGAINS

Corydon, Indiana
September 6, 1904

Olney, Maryland
November 1, 1981

Mr. Robert A. Ragains was interested, chiefly, in Entomology and was retired from 34 years of service with the United States Department of Agriculture Home Administration where he was a Credit Specialist. Some biographic material was found in the *Thomas Say Alumnus 1972-1973*, Department of Entomology, Purdue University supplied to us by Mr. John Favinger, a friend of Mr. Ragains while in Indiana. Mrs. Ragains also provided material.

Mr. Ragains was born at Corydon, Indiana, and attended public school in Hardinsburg, Indiana. He then studied at Indiana Central University where he obtained an A.B. degree in 1928. He majored in Entomology at Purdue University where he earned a Master of Science degree in 1933.

Mr. Ragains taught 4 years in high school at Zionsville, Indiana before employment by the United States Department of Agriculture. He was living in Zionsville in 1931 when he joined the Indiana Academy of Science. When at Purdue University, Mr. Ragains wrote a thesis (unpublished) including a list of ground beetles which, according to John Favinger, Indiana State Entomologist, has been very useful to him. Mr. Ragains was not in Indiana long enough after joining to be very active in the Academy, but must have valued the association highly, because he was still a Senior Member at death.

Unfortunately, biographical material about Mr. Ragains seemed meagre; and as he was out-of-state for so long many of us did not know him. His brief note to the *Thomas Say Alumnus* is very revealing, though. In that, his hobbies of golfing, woodworking and raising azaleas are mentioned as retirement activities. Three grandchildren are also mentioned as "really Grand and the joys of our lives".

Mr. Ragains was 77 years old when he died suddenly of a heart attack November 1, 1981, in a hospital in Olney, Maryland.

BERNARD HENRY SCHOCKEL

Aurora, Indiana
December 15, 1887

Lawrenceburg, Indiana
March 5, 1972

Dr. Bernard Henry Schockel was interested chiefly in Geography and Geology. Born at Aurora, Indiana, December 15, 1887, his early education was secured locally according to a record of training and professional experience kindly supplied to us by the archivist at Valparaiso University where Dr. Schockel taught as a visiting lecturer from 1954 to 1957. Dr. Schockel graduated from high school at Aurora June, 1905, and began teaching in a country school of Washington Township that year. The next year, he entered Indiana State Normal where he received a diploma in 1909. He received a B.S. degree from the University of Chicago in 1911 and an M.A. in 1913 and Ph. D. degree in 1947. His major was Geography and Geology. He did a survey of the Galena, Illinois, Quadrangle and later wrote a bulletin for the Illinois Geological Survey. The survey was done in 1910.

In 1913, Dr. Schockel was elected head of the Department of Geography and Geology by the board of Indiana State Teachers College. In the *History of Indiana State Teachers College 1865-1945* by William O. Lynch (material by courtesy of the Public Relations Office of Indiana State University), Dr. Schockel is described as "a young man of much promise already on the road to high scholarship". He was employed after the resignation of Dr. Charles Dryer, a charter member of the Indiana Academy of Science.

When World War I began, he had a leave of absence to enter Meteorological School of the American Expeditionary Force at Langres, France, in preparation for service in the U.S. Signal Corps. He served well receiving a military citation.

Dr. Schockel returned from France to Indiana State, but had numerous leaves of absence to further graduate study and he carried on concurrent work to help with finances. He was employed by the U.S. Weather Bureau, St. Louis, Missouri in 1917, working consecutively in every position up to and including forecasting. He was a special expert with the U.S. Shipping Board (Planning and Statistics) 1919 to 1920. In the 1920s, he conducted field trips for adults to the East Coast and Rocky Mountains. He had about 7 classes in Geology and Geography at the University of Chicago interspersed from 1912 to 1936, which he conducted.

When Dr. Schockel asked for an extension of a leave of absence from Indiana State from 1931 to 1932, he was notified of the termination of his service after the 1932 to 1933 year, because it had become necessary to reduce the staff in his field.

He had another class at the University of Chicago in 1932 and was special expert with the Tennessee Valley Authority (Planning and Housing Div.) from 1934 to 1935 making land use studies and surveys of the area. From 1935 to 1936, he was head of the Geography and Geology Department of YMCA Central College, now Roosevelt College, Chicago, Illinois. He took classes at the University of Chicago in 1934 and 1936. Concurrent with all of the other activities, Dr. Schockel spent 20 years in newspaper work from 1907 to 1954. He contributed hundreds of newspaper articles to the *Terre Haute Star, Indianapolis Star, Chicago-Record Herald* and others. He was visiting Geography Professor at Washington University from 1947 to 1948 and also at the University of Kentucky from 1951 to 1952. Meanwhile, he spent 3 years at Mankato Teachers College, Minnesota, 1948 to 1949, 1949 to 1950, and 1952 to 1953. In 1952, he spent a month on the Pacific Coast, and in 1953 he stayed a month on the East Coast and visited Canada.

As mentioned before, Dr. Schockel was a visiting lecturer in the Department of Geography and Geology at Valparaiso University, Valparaiso, Indiana from 1954 to 1957.

Dr. Schockel joined the Indiana Academy of Science in 1913 and became a Fellow in 1917. He was one of the few Life Members of the Society. He was Chairman of the Geology and Geography Section in 1956, and gave about 17 papers at Academy meetings. Some of his chief work consists of the book, *Manufactural Evansville* (submitted in candidacy for the Ph. D. degree), *Pictorial Map of Indiana* (1924), *Indiana Supplement for the Atwood Geography, Changing Conditions in the Kentucky Mountains* (Scientific Monthly), *Pupils Help Book in Geography*. He also originated three dimensional maps in black and white or color, produced a land use map of the Mediterranean, studied land use in Indiana and estimated the economic importance of Indiana's counties. In addition to membership in the Academy, Dr. Schockel was a member of the Association of American Geographers, Indiana Historical Society, Sigma Xi, Phi Kappa Sigma, Legion and Veterans of Foreign Wars.

After retiring from teaching at Valparaiso in 1957, Dr. Schockel wrote articles for the local newspaper about problems concerning the locality, Aurora, Indiana, where he was living. He was an avid reader, gardener and sportsman according to his niece, Elizabeth Rullman. He played golf up until a few months before death at 85 years of age. His physical-political map of Indiana was the official one until asked to prepare a new one just before he died. His age and health would not permit it. He died in the Dearborn County Hospital, Lawrenceburg, Indiana, March 5, 1972. We regret that we learned of his death only recently.

WILLIAM JOHN TINKLE

Grant County near Marion, Indiana — North Manchester, Indiana
November 20, 1892 — June 12, 1981

Dr. William John Tinkle was a biologist with a long teaching record in Indiana. Born in Grant County near Marion, Indiana, November 20, 1892, he was educated in district and high schools at Sweetser until 1911. From 1911 to 1913, he taught school at Sweetser, Indiana. He went to Manchester College, North Manchester, Indiana, where he secured an A.B. degree in 1916. We are indebted to the *History of the Mississinewa Church of the Brethern* by Ralph G. Rarick, 1917, for a glimpse of his early years. Dr. Tinkle was ordained a minister in 1914 at Somerset. He soon placed membership with the Mississinewa Church, but was absent so much because of the academic work at Manchester College that he could devote little time to the pulpit. He was married in August, 1916, and a few days later, moved to Taft, Texas, where he and his wife taught and engaged in church work in a pioneer settlement. In 1922 to 1923, Dr. Tinkle was a lecturer at the World's Purity Federation and was back to teaching high school in Indiana from 1924 to 1926. Interspersed with the teaching, he worked on an M.A. degree at Ohio State University receiving the degree in 1927. He was a Professor of Natural Science at Mission House College from 1927 to 1929 and Head of the Department of Biology at Juniata College from 1929 to 1931. He had classes at Wisconsin in 1929. He returned in earnest to his education and was an assistant in Zoology at Ohio State University from 1931 to 1932, and received a Ph. D. degree in 1932. After graduation, he taught a year at Marshall College, but moved to Taylor University at Upland, Indiana in 1933 as an Instructor in Biology. He moved up to professor by the 1939-1940 school year. From 1941 to 1944, he was Professor and Head of the Department of Biology, La Verne College. He was back at Taylor University as Chairman of the Division of Science from 1944 to 1948. From 1946 to 1949, he spent summers teaching at Ball State University and was an associate professor at Anderson College from 1956 to 1958 when he retired.

Dr. Tinkle joined the Indiana Academy of Science in 1934 while at Taylor University. He was on the Program Committee in 1936, the Membership Committee from 1936 to 1939 and was Divisional Chairman of the History of Science Division in 1956. He gave several papers at annual meetings and wrote a memorial for Hal Herbert Coffel. He spoke to the Academy on the genetics of sweet peas, Darwinism, conservation of the germ plasm of species, biological theories of Henri Fabre, effectiveness of natural selection, and Creationism. He was also a member of the Genetics Association and American Scientific Affiliation. He is listed in *American Men of Science* and *Indiana Scientists* by Visher. Dr. Tinkle lived in Eaton, Indiana, before entering Timbercrest Church of the Brethern Retirement Home at North Manchester, Indiana, January 18, 1973. His last paper before the Academy, the one on Creationism in the 20th century was given that fall. At 88 years of age, Dr. Tinkle died, June 12, 1981, at the Timbercrest Health Care facilities, and was buried at Eaton, Indiana. We are indebted to Wanda E. Miller, Assistant Administrator of Timbercrest, and Betty G. Freese, Associate Director of Alumni Relations at Taylor University for information about him.

YERAM SARKIS TOULOUKIAN

Istanbul, Turkey
December 28, 1920

Bethesda, Maryland
June 12, 1981

Dr. Yeram Sarkis Touloukian was a world renowned mechanical engineer specializing in the thermophysical properties of substances. He was born in Istanbul, Turkey, December 28, 1920, to an Armenian merchant family. He studied there at Robert College of Engineering receiving a B.S.M.E. degree in 1939. He came to the United States of America to attend the Massachusetts Institute of Technology on a tuition scholarship receiving an M.S. M.E. degree in 1941. He was an assistant and instructor in Mechanical Engineering there from 1941 to 1943. In 1944, he went to Purdue University on a teaching fellowship and studied under G.A. Hawkins toward a Ph. D. degree majoring in heat transfer. He received the doctorate in 1946. He became an assistant professor in 1947, associate professor in 1953 and professor in 1957. He had become an American citizen in 1952.

Dr. Touloukian and his work were highly praised in a Purdue University *Thermophysics and Electronics Newsletter* (V. 10, No. 3) dedicated to him and some issues of the *Purdue Alumnus* which were kindly sent to us by Dr. W.M. Phillips, Head of the Purdue University School of Mechanical Engineering. Dr. Touloukian received the Alcoa Distinguished Professor of Engineering Award in 1967 at Purdue; and in 1968, the Gold Medal of the Italian National Society of Thermal Engineering in ceremonies at Bologne, Italy. In 1970, he was honored by Purdue again by election to the chair of Distinguished Atkins Professor of Engineering. He received the American Society of Mechanical Engineers Heat Transfer "Memorial Award" in 1974; the International Thermal Conductivity Conferences "Thermal Conductivity Award" in 1977; and the International Thermophysics Congress "Distinguished Achievement Award" in 1981. He was invited by the U.S.S.R. Academy of Sciences to conduct a lecture tour of their country in April to May, 1967.

Professor Touloukian taught numerous courses at Purdue in heat and mass transfer and developed many new courses in physical thermodynamics. Many of his students became prominent in this country and others. He was the major professor of about 65 Ph. D. and M.S. degree candidates.

Another outstanding achievement was the conception, founding and implementing of the Thermophysical Properties Research Center (TPRC) at Purdue University in 1957. Its purpose was the synthesis and reasearch of world knowledge of the thermophysical properties of materials vitally important to science, engineering and technology. According to the memorial newsletter article by C.Y. Ho, it attained national and international recognition in a very short time. It was cited by President Eisenhower's Science Advisory Committee in 1958 as an example of centers offering a solution to acute problems of scientific and technical information needs of the nation. Dr. Alvin Weinberg, author of the *Weinberg Report*, from the U.S. President's Science Advisory Committee cited the Purdue research center as a model data analysis center. In 1960, the Air Force had established the Department of Defense Thermophysical Properties Information Center (TPIC) at Purdue's TPR Center. Other operations joined in rapid succession: in 1964, an external center for the National Bureau of Standards, a component of the National Standard Reference Data System (NSRDS); in 1972, the Underground Excavation and Rock Properties Information Center (UERPIC) established by the National Science Foundation; in 1973, the Electronic Properties Information Center (EPIC) moved to Purdue by the Defense Logistics Agency from Hughes

Aircraft to merge with TPIC. The latter move resulted in establishing the Department of Defense Thermophysical and Electronic Properties Information and Analysis Center (TEPIAC) of Purdue's TPRC.

After 17 short years in 1974, the center was reorganized becoming the Center for Information and Numerical Data Analysis (CINDAS) which incorporated all of the operations. The Department of Defense and National Bureau of Standards have long recognized the center at Purdue as a national center for thermophysical and electronic properties data. Dr. Ho states, "The steady and continued growth of CINDAS fully testifies to Dr. Touloukian's outstanding technical and administrative capabilities. Furthermore, his sustained and untiring insistence for the need of such data centers as CINDAS is now fully supported and echoed by many responsible bodies." CINDAS is cited as an example to follow to meet a national crisis in the "information explosion". Dr. Touloukian's counsel and views were sought by many national and international scientific and technical societies as well as industry and governmental agencies. He served on many committees and governmental panels and held several patents.

Dr. Touloukian joined the Indiana Academy of Science in 1951 when teaching in the School of Mechanical Engineering at Purdue University. He was an author or co-author of papers given at meetings on design of a sensitive dew point instrument and electrical analog circuits as applied to heat conduction problems. He was a Regular Member. He was Fellow of the American Society of Mechanical Engineers and American Institute of Aeronautics and Astronautics; and a member of the American Association for the Advancement of Science, American Society of Information Science, Pi Tau Sigma Mechanical Engineering Honorary Fraternity, Sigma Xi Scientific Research Society (Pres., Purdue Chap., 1975-76). A memorial fund has been established at Purdue in his honor. He can be found in *Who's Who in Indiana* (Hist. Record Assoc.), *Who's Who in the Midwest* (V. 8, Marquis), and *Indiana Scientists* by Visher.

He was attending one of his many meetings when he suffered a massive heart attack and died two hours later. He was at Bethesda, Maryland, June 12, 1981, when the attack occurred at a meeting of an international task group on Thermophysical Properties. In addition to other functions to perform at that meeting, he was also to deliver a welcoming speech at the National Bureau of Standards First International Joint Conferences on Thermophysical Properties and a banquet speech June 16. He was to be presented with a surprise distinguished achievement award at that banquet. He was only 60 years of age when the scientific community was shocked by his sudden death.

For further details of the many interests and accomplishments of this vigorous and enthusiastic man, a reading of the May/June, 1981 (V. 8, No. 3) *Thermophysics and Electronic Newsletter* cited above is highly recommended. In addition to his scientific achievements, he was a warm, happy individual, family man, good host and interested in young people. He was an avid collector of mementos from his wide travels.

INDIANA ACADEMY SCIENCE
NEW MEMBERS 1982

ALSOP, DR. TED. J., Dept. of Geography and Geology, Indiana State University, Terre Haute, IN. 47809

ARGOT, DR. JEANNE, Marion College, Marion, IN 46952

AUSLINGER, MR. C. MICHAEL, Anthropology Laboratory, Indiana State University, Terre Haute, IN 47809

BAKER, MR. TOM, Peru High School, 401 N. Broadway, Peru, IN 46970

BARBER, MRS. DIANE, Dept. of Biology, IU-PU at Fort Wayne, 2101 Coliseum Blvd. E., Fort Wayne, IN 46805

BASU, DR. ABHIJIT, Dept. of Geology, Indiana University, Bloomington, IN 47405

BECKMAN, DR. JEAN C., Dept. of Chemistry, University of Evansville, Evansville, IN 47702

BENWAY, MS. DENISE, 1509 S. Macedonia Avenue, Muncie, IN 47302

BEYNON, DR. DIANE E., IU-PU at Fort Wayne, Fort Wayne, IN 46805

BLEDSOE, MR. LARRY W., Dept. of Entomology, Purdue University, West Lafayette, IN 47907

BROWN, MR. RICHARD B., 2424 E. Kessler Blvd., Indianapolis, IN 46220

BURNETT, DR. J. PAUL, Lilly Research Laboratories, 307 E. McCarty Street, Indianapolis, IN 46285

CAPOUCH, MR. BRIAN, R.R. #1, Box 270, Monon, IN 47959

CAPSHEW, MR. JAMES H., Dept. of History and Sociology of Science, Smith Hall/D6, University of Pennsylvania, Philadelphia, PA 19104

CARTER, MS. CHARLOTTE, Dept. of Chemistry, Purdue University, West Lafayette, IN 47907

CASTRALE, DR. JOHN S., Forest Wildlife Office, R.R. #2, Box 477, Mitchell, IN 47446

CHIANG, DR. PEN-CHI, Dept. of Civil Eng., Purdue University, West Lafayette, IN 47906

CUDMORE, MR. WYNN W., Dept. of Life Sciences, Indiana State University, Terre Haute, IN 47809

CUNDIFF, MS. GEORGINA L., 1600 Harmony Way, Evansville, IN 47712

CURRAN, MR. JOHN T., National Weather Service, Indianapolis International Airport, Indianapolis, IN 46251

DAVIS, DR. GRAYSON S., Dept. of Biology, Valparaiso University, Valparaiso, IN 46383

DEMANNO, MISS DEBORAH A., Dept. of Biology, University of Notre Dame, Notre Dame, IN 46556

DHAWALE, DR. SHRIKRISHNA W., Indiana University East, Richmond, IN 47374

DROSTE, DR. JOHN B., Dept. of Geology, Indiana University, Bloomington, IN 47405

FARON, MR. EUGENE J. II, 9317 Foliage Lane, Munster, IN 46321

FRANKLIN, DR. THOMAS D. JR., Indiana University Hospital, A-32, 1100 W. Michigan, Indianapolis, IN 46223

GARCIA, DR. J.R., Dept. of Biology, Ball State University, Muncie, IN 47306

GARCIA-HERNANDEZ, MS. ALICIA, 3710 Green Ash Ct., Indianapolis, IN 46222

GHOSE, MR. S.N., Amax Coal Company, 105 S. Meridian Street, P.O. Box 967, Indianapolis, IN 46206

GOEBEL, DR. EDWIN M., Dept. of Biological Sciences, IU-PU at Fort Wayne, 2102 Coliseum Blvd. E., Fort Wayne, IN 46805

GONCZGROW, MR. ANTHONY T., 138-004 Nimitz Dr., West Lafayette, IN 47906

GROSS, MR. WILLIAM H., Dept. of Chemistry, Ball State University, Muncie, IN 47306

HANLEY, DR. THOMAS R., Rose-Hulman Inst. of Technology, Terre Haute, IN 47803

HARRIS, DR. RONALD L., IU-PU at Fort Wayne, 2102 Coliseum Blvd., E., Fort Wayne, IN 46805

HAZEL, DR. WADE N., Dept. of Zoology, Depauw University, Greencastle, IN 46135

HERRING, MRS. JANICE R., Lilly Research Laboratories, Dept. 626, 307 East McCarty Street, Indianapolis, IN 46285

HINES, MR. DICK, Dept. of Biology, Ball State University, Muncie, IN 47306

HOLDAWAY, MR. PAUL A., Harper Community College, Palatine, IL 60067

HOLMES, MR. JAMES. E., 521 South Park #2, Bloomington, IN 47401

HOSWATH, DR. KATHLEEN L., Dept. of Biology, Notre Dame, IN 46556

HUGHES, MR. AUSTIN L., Dept. of Biology, Indiana University, Bloomington, IN 47405

HOMOYA, MR. MICHAEL A., Indiana Natural Heritage Program, Rm 612 State Office Building, Indianapolis, IN 46204

HUGHES, MRS. MARY ANN, Dept. of Biology, Indiana University, Bloomington, IN 47405

JAMES, MR. DAVID, WRTV, 1330 N. Meridian, Indianapolis, IN 46206

JOHNSON, DR. SUSAN M., Dept. of Biology, Ball State University, Muncie, IN 47306

KAMO, MS. KATHRYN, Dept. of Biology, Indiana University, Bloomington, IN 47405

KARAMOUZ, MR. MOHAMMAD, School of Civil Engineering, Purdue University, West Lafayette, IN 47907

KELLER, DR. CLIFTON, Andrews University, Berrien Center, MI 49104

KINKADE, MS. KRISTEN E., 714 S. Cole Ave. #32, Muncie, IN 47303

KLADIVKO, DR. EILEEN J., Dept. of Agronomy, Purdue University, West Lafayette, IN 47907

KLEINHANS, DR. FREDERICK W., IUPUI, P.O. Box 647, Indianapolis, IN 46223

KRAMER, MS. MARGARETA H., Dept. of Biology, Indiana University, Bloomington, IN 47405

KRUGER, DR. TERRY L., Dept. of Chemistry, Ball State University, Muncie, IN 47306

LANGELL, MR. GARY L., Division of Fish & Wildlife, 4201 E. 3rd St., Bloomington, IN 47401

LATZ, KATHLEEN, 1337 West Wayne Street, Fort Wayne, IN 46804

LOVVORN, MR. JAMES R., School of Natural Resources, University of Michigan, Ann Arbor, MI 48109

LULLA, D. KAMLESH, Dept. of Geography and Geology, Indiana State University, Remote Sensing Laboratory, Terre Haute, IN 47809

MCCRARY, MS. KAY, Walnut Grove Trailers #185, Bloomington, IN 47401

MCCREARY, MS. NANCY. J., Box 17 Galvin Life Sciences, Dept. of Biology, University of Notre Dame, Notre Dame, IN 46556

MANGOLD, MR. WILLIAM L., Resource Analysts, P.O. Box 2477, Bloomington, IN 47402

MARWAHA, DR. JOE, I.U. School of Medicine, T.H.C.M.E., 135 Holmstedt Hall, Terre Haute, IN 47809

MAUPIN, MR. MARTY, 2765 Lawson Rd., Marion, IN 46952

MEISS, DR. RICHARD A., I.U. School of Medicine, 102A Medical Research Facil, 1100 W. Michigan St., Indianapolis, IN 46223

MESS, MR. JOHN. A., 5129, G.R.C., Bloomington, IN 47401

MIANO, MR. JOE, St. Joseph's High, 1441 No. Michigan St., South Bend, IN 46617

MILLER, DR. JAMES S., Goshen College, Goshen, IN 46526

MILLER, MR. M. ERIC, Goshen College, Goshen, IN 46526

MINTON, MS. MADGE R., 4840 E. 77th St., Indianapolis, IN 46250
MITCHELL. MR. DOUGLAS R., Indiana Dept. of Highways, Rm. 1204 State Office Bldg., 100 N. Senate Ave., Indianapolis, IN 46204
MIZELL, DR. SHERWIN, Medical Sciences Program, Indiana University, Bloomington, IN 47405
MOORE, MRS. KATHRYN, 7234 Hosler Rd., Leo, IN 46765
MORIARTY, MR. JOSEPH A., John Adams High School, 808 S. Twyckenham Dr., South Bend, IN 46615
MORRIS, MR. TIMOTHY J., West Middle School, 109 E. Garfield, Martinsville, IN 46151
MORRISON, DR. WILLIAM A., Dept. of Chemistry, University of Evansville, Evansville, IN 47702
MUNFORD, MR. JOHN W., Dept. of Biology, Wabash College, Crawfordsville, IN 47933
NELSON, MR. CRAIG A., Dept. of Biology, Ball State University, Muncie, IN 47306
NOWAK, DR. THOMAS, Dept. of Chemistry, University of Notre Dame, Notre Dame, IN 46556
OCHS, DR. SIDNEY, Dept. of Physiology, I.U. School of Medicine, Indianapolis, IN 46223
OLSEN, DR. ROBERT J., Dept. of Chemistry, Wabash College, Crawfordsville, IN 47933
OLSEN, DR. JULIE A., Dept. of Chemistry, Wabash College, Crawfordsville, IN 47933
O'NELL, DR. CARL W., Dept. of Anthropology, University of Notre Dame, Notre Dame, IN 46556
O'NELL, DR. NANCY D., Purdue University, North Central, Westville, IN 46391
PENNINGTON, DR. JODIE A., Dept. of Animal Science, Purdue University, West Lafayette, IN 47907
PISTOLE, MR. DAVID H., Dept. of Life Sciences, Indiana State University, Terre Haute, IN 47809
PLEW, MR. JIM, Engineering Aggregates Corp., P.O. Bos 538, Logansport, IN 46947
POHLEY, DR. WILLIAM J., Dept. of Biology, Franklin College, Franklin, IN 46131
PRZYBYLINSKI, MR. RON, National Weather Service Forecast Office, Indianapolis International Airport, P.O. Box 51526, Indianapolis, IN 46224
REED, DR. GARY L., Fruit and Vegetable Insects Research, U.S.D.A., A.R.S., P.O. Box 944, Vincennes, IN 47591
RESEIGH, MRS. MELANIE, 1906 West 11th St., Muncie, IN 47302
RESEIGH, MR. WM. E., Dept. of Anthropology, Lucina Hall, Ball State University, Muncie, IN 47306
RHODES, DR. SAMUEL B., Dept. of Biology, Franklin College, Franklin, IN 46131
RICHARDS, MR. DAVID D., Creston Junior High, 10925 E. Prospect St., Indianapolis, IN 46234
ROBERTS, MS. MARLINE A., Box 1595B, Indianapolis, IN 46206
ROBERTSON, MR. ROBERT N., Bass Lake State Fish Hatchery, R. 3, Box 240, Knox, IN 46534
RYBARCZYK, DR. JAMES P., Dept. of Chemistry, Ball State University, Muncie, IN 47306
SALERNO, MR. BARRY J., Ball State University, Cooper Life Science Bldg., Muncie, IN 47306
SASSAN, MESHGIN-AZARIAN, 277 Eigenmann Hall, Indiana University, Bloomington, IN 47406
SCHOKNECHT, DR. JEAN, Indiana State University, Terre Haute, IN 47809
SKINNER, MR. JERRY M., I.S.U.E., 8600 University Blvd., Evansville, IN 47712

SMITH, MS. MARTHA, Dept. of Life Science, Indiana State University, Terre Haute, IN 47809
SMITH, MR. MICHAEL D., 1415 North Jordan Ave., Bloomington, IN 47401
SNYDER, MR. JOSEPH H., 1130 W. Michigan St., Indianapolis, IN 46206
STIER, MR. STEVEN L., 8712 Notestine Rd., Ft. Wayne, IN 46815
STILLWELL, DR. WILLIAM, Dept. of Biology, I.U.P.U.I., 1125 E. 38th St., Indianapolis, IN 46223
STITSWORTH, MR. MICHAEL H., AGAD 228, Purdue University, West Lafayette, IN 47907
STONE, LEE ANN, Ball Corporation, 1509 S. Macedonia, Muncie, IN 47302
SWIERGIEL, MS. JENNIFER J., Ashton-Aley M262, Bloomington, IN 47406
TAYLOR, DR. MILTON W., Dept. of Biology, Indiana University, Bloomington, IN 47402
THIEL, MR. DANIEL P., Anthropology Laboratory, Indiana State University, Terre Haute, IN 47809
TRUMPEY, MR. JERRY, 4879 Spring Grove Ave., Cincinnati, OH 45232
TUCKFIELD, MR. R. CARY, Dept. of Biology, JH 105, Indiana University, Bloomington, IN 47405
VANTYLE, DR. W. KENT, 4600 Sunset Avenue, Indianapolis, IN 46208
WALTON, MR. RODNEY L., Jordan Hall A64, Bloomington, IN 47405
WATT, MS. DORIS, Dept. of Biology, St. Mary's College, Notre, Dame, IN 46556
WEATHERHOLT, MR. RAY JR., R. 2, Box 445A, New Albany, IN 47150
WIDLAK, DR. FREDERIC W., 222 E. Ohio St., Indianapolis, IN 46204
WILKINSON, MR. ANTHONY, 612 State Office Building, Indianapolis, IN 46204
YATSKIEVYCH, MR. GEORGE, Dept. of Biology, Indiana University, Bloomington, IN 47405
YEAGLEY, MS. THERESE, Western Kentucky University, P.O. Box 801 Bemis-Lawrence Hall, Bowling Green, KY 42101
HAYES, MRS. CAROLYN A., Center Grove High School, 2717 S. Morgantown, Greenwood, IN 46142
SOUTH VIGO JETS CLUB, c/o B.J. Ridens, 3737 So. 7th, Terre Haute, IN 47802
INDIANA STATE MUSEUM, 202 N. Alabama St., Indianapolis, IN 46204

INDIANA SCIENCE TALENT SEARCH HONORS LUNCHEON

Indianapolis

March 13, 1982

Dr. William Eberly, President of the Indiana Acacemy of Science and Professor of Biology at Manchester College, congratulates Richard Meyer for being named one of two $1,000 scholarship winners in the 1981-82 Indiana Science Talent Search. Richard, a senior at East Noble High School in Kendallville, plans to study horticulture in college and then manage his family's orchard.

These scholarships are provided by Kappa Kappa Kappa Sorority.

ADDRESS AND CONTRIBUTED PAPERS

CREATIONISM AND EVOLUTION: SOME PERSPECTIVES

WILLIAM R. EBERLY
Department of Biology, Manchester College,
North Manchester, Indiana 46962

All of us have heard about the famous trial at Dayton, Tennessee, of the young biology teacher who dared to say a few words about evolution in the high school. After a lapse of several decades, there is again considerable interest in the subject of evolution and creationism. There has been another flurry of legislative action, law suits, books, pamphlets, and radio and television presentations against evolution. A few years back, the state of California passed a law which required that creationism be taught in the public schools. One book publishing company had a very popular textbook in biology in which the section on human origins included a picture of the British paleontologist, Dr. Leakey. In a special edition of the textbook prepared for sale in California, the publishers replaced that picture with a picture of Michelangelo's Sistine Chapel fresco of the creation of man. The source of this bit of information said that the subsititution of Adam for Leakey symbolizes the two sides of this controversy over the teaching of science in the public schools. (23) It is a movement, a dialogue, a controversy which has swept the country in the last several decades.

Creationism is essentially an expression of a religious point of view. My own involvement in religious activities and scientific research over many years has given me an opportunity to assess both sides of this question.[1] And I thought perhaps some of my ideas and interpretations might be of interest to some of you. I have no intention of saying anything that would insult or antagonize anyone. What I say I want to say as kindly as I can and hopefully with as much support and background as I can present.

In order to understand creationism, we need to look into the history and the background of this idea. A recent book on church history included this statement, "Many Christians today suffer from historical amnesia. The time between the apostles and their own day is one great blank. As a consequence of our ignorance concerning Christian history, we find believers vulnerable to the appeals of cultists. Some distortion of Christianity is often taken for the real thing". (20, p. 9)

In the middle of the 19th century there were four main streams of scientific and cultural development converging at the time of Charles Darwin. The first of these was the culmination of a long period of scientific research where discoveries during the preceeding three or four centuries changed completely our conception of the world. We recall such names as Copernicus, Kepler, Galileo, Newton and Lyell. We know how they changed our ideas of astronomy and geology and physics. To all of these revelations, the church reacted vehemenently, imprisoning people, punishing them, persecuting and torturing them. The second

1. In addition to his work as a scientist and teacher, Dr. Eberly has been a minister in the Church of the Brethren since 1943. He has served as a pastor and counselor and has filled many administrative posts from the local level to the national denomination. For 10 years he was the secretary of the Annual Conference, the national legislative body, and 1980 served as the Moderator, the highest elected office in the denomination.

of the great trends was the development of our understanding of the ideas of evolution. Many people seem to credit Charles Darwin with inventing evolution. But the idea of evolution, the concept of change, goes back hundreds of years. Some even trace it back to the ancient Greeks. A recent book listed 24 published authors who conceived of the idea of evolution as applied to the origin of species before 1859, a century or so before the time of Darwin. (11, p. 133) The idea of evolution, we could almost say the necessity of evolution as an idea, was well known to the scientific and intellectual community. But there was lacking the kind of data and information which would help explain the mechanisms of evolution and therefore the idea was pretty much an abstraction.

The third trend emerging at this time in history was the result of the scholarly study of the Bible within the theological community. There were new approaches to the study of the Bible. Scholars were beginning to study the Bible as they would any historical document. This included close scrutiny of the manuscript text, the language, the contemporary culture, and so on. The name given to this general approach is Higher Criticism. Higher Criticism of the Bible had been developing over several hundred years and was giving people a whole new concept of what the Bible is and how it is to be interpreted. (22) The fourth trend was unique to the United States. In the middle of the 19th century we were struggling with slavery and were trying to decide whether we wanted to perpetuate the institution of slavery or not. Those who supported slavery found support in the Bible. Those who opposed slavery likewise took their cue from the Bible. The North won; the South lost. This had a significant impact on the subsequent religious development in America in the division of the United States into a northern and a southern theology of the Bible. Andrew White, in his classic *History of the Warfare of Science and Theology* says, "Nothing during the American struggle against the slave system did more to wean religious and God fearing men and women from the old interpretation of scripture than the use of it to justify slavery". (24, ii, p. 368)

Into this explosive mixture of ideas and trends came Darwin's book, *The Origin of Species*, on November 24, 1859. All of the 1,250 copies were sold out the first day. Darwin's book on evolution made plausible and possible that which before seemed to be a complete abstraction.

Darwinism was seen by some to be a threat to religion and the authority of the church. The reaction in England was immediate and rather intense. In the United States, the civil war delayed the public response for several decades, but inevitably it came. There was a big conference of evangelical church leaders in New York in 1873. During this "Evangelical Alliance" there were several sessions of papers and speeches on "Christianity and its antagonists". There were a number of papers on Darwinism and the effect that this would have on the church. And there was considerable debate, much of it unscheduled.

The participants at this assembly soon began to move into two different camps in terms of their reaction and response to the impact of Darwinism and everything that went with it. One group seemed to say that the discoveries of science are inevitable. We've faced this before and the church must recognize truth wherever it finds it. The church ought to concentrate its efforts on individuals, their personal life, the relationships of people. The most eloquent speaker for this faction was Henry Ward Beecher. Those who took that general point were thereafter referred to as liberals.

There were others at the conference who felt that they were giving up too much and that they shouldn't sacrifice their great Biblical truths. They felt that

evolution, and Darwinism specifically, was the enemy of the church and that loyal Christians should rise up and combat it. The chief spokesmen for this point of view were Jonathon Blanchard, president of Wheaton College, and Dwight L. Moody, popular evangelist of that day. Those who took this stance could be referred to as conservatives.

Now at this point in church history there were also several other very active movements. There was a wide-spread interest in prophecy. There were the millenarists who looked forward to an early, bodily return of Christ to the earth. There was the Sunday School movement in the 19th century. And there was the Revival movement. So a lot of things were happening. Many of the more conservative persons from various of these movements began gradually to separate themselves more and more from the main line church members and began to have their own conferences. One such conference, referred to as the Niagara Bible Conference, in 1890 adopted a series of 14 planks or statements which became their creed, their statement of faith. The *first* of these 14 planks in 1890 was a statement affirming the literal, word for word, inerrant inspiration and transcription of the Bible. (13)

This represented a significant but very subtle change in the views that people had held before about the Bible. It was not unusual up to this time to find many people who would say, 'yes, I believe the Bible. I believe it literally. I believe it word for word'. But they were describing the mechanism of their own faith. This new point of view seemed to ascribe some characters to the book itself. It seemed to affirm some new properties of the Bible quite separate from the point of view of anyone reading it.

In 1910 there appeared a publication, a paperback booklet called *The Fundamentals of the Faith*, that was followed by 11 more during a 5 year period. (2) These books, *The Fundamentals*, were sent free and unrequested to all protestant ministers in the United States. The project was financed by a couple of wealthy businessmen. The people who gathered around the ideas in these books began in 1920 to refer to themselves as "fundamentalists", while others began to call the movement "fundamentalism".

By this time the fundamentalists' platform had been reduced to about 4 or 5 major points, the first one of which was always an affirmation of the literal, inerrant form of the Bible. Inevitably, scientists who belonged to the fundamentalist movement began to be aware of some of the dilemmas of their situation. So in 1941 there was organized the American Scientific Affiliation. In order to belong to the ASA, a scientist had to commit himself to the following statement or creed (in part): "I believe that the Bible is uniquely inspired of God, is inerrant in the original writings, and that it gives me a true revelation of God. Since God is the Author of the book, as well as the Creator and Sustainer of the physical world about us, I believe there can be no discrepancy between biblical statements and scientific observations when both are properly interpreted. . . .I consider the present text of the Bible to give reliable evidence of the exact words of the original Scripture save where clear manuscript evidence shows that an error of transmission has occurred. This Bible I accept as my final authority in matters of faith and conduct". (1)

In 1963, another organization arose called the Creation Research Society. Members have to commit themselves to this statement: "1. The Bible is the written word of God, and because it is inspired throughout, all its assertions are historically and scientifically true in all the original autographs. To the student of nature, this means that the account of origins in Genesis is a factual presenta-

tion of simple historical truths. 2. All basic types of living things, including man, were made by direct acts of God during the Creation Week described in Genesis. Whatever biological changes have occurred since Creation Week have accomplished only changes within the original kind". (14, p. 608)

And so a scientist within the fundamentalist movement commits him/herself first of all to accept the literal interpretation of the Bible and then tries to fit all scientific data and discoveries into that system. Henry M. Morris, one of the chief spokesmen of the creationist movement today, says point blank, "There is not the slightest possibility that the facts of science can contradict the Bible". (16, p. 15)

Duane Gish, the creationists' chief authority on fossils, says that man and the dinosaurs were created together and lived together on the earth at the same time. How do we know this? The Bible says so, says Gish. "The Bible tells us that God made man and the dinosaurs (included among the beasts of the earth) on the 6th day of creation". (9, p. 13)

So firm is this belief in the literal inspiration of the whole Bible that Kenneth Taylor would say, "If the Bible is not true about creation, then what parts of scripture are true, and how can one decide which are true and which are not". (21, p. 7)

It is interesting to note that Darwin, speaking of himself during his seminary days (and he was in the seminary and planned at one time to become a minister), said, "I did not then in the least doubt the strict and literal truth of every word in the Bible". (5, i, p. 39)

It should be pointed out that the idea of verbal inspiration and literal inerrancy of the Bible is by no means universal among Christians or among church groups in general. It might be observed, for example, that the Bible was not available to the members of the church for nearly three quarters of the entire existence of the church. Until the invention of printing, only a few scholars in the monasteries and some priests had access to the Bible. There is no particular internal evidence, that is, scriptures within the Bible, that speaks to this point and supports this particular concept. In fact, just the opposite is true, in my judgement. The New Testament speaks many times about God revealing himself through a person, Jesus, and never through a printed book. In fact, the phrase "Word of God" as used in the Gospel of John (chapter 1) referred to Jesus, a living, walking, talking human being, and not to a written, printed word.

The New Testament records numerous confrontations between Jesus and the leaders of the early church who insisted on a literal emphasis of *their* scripture (the Old Testament) to the exclusion of all other sources of religious truth, even the Son of God himself. In the Sermon on the Mount (Matthew, chapter 5-7), Jesus, in reference to at least six different subjects, said, "You have heard that it was said of old. . . . but I say to you . . . " On another occasion, Jesus was confronting the fundamentalists of his day, when he told them bluntly, "You search the scriptures, for you believe *they* give you eternal life, and the scriptures point to me. Yet you won't come to me so that I can give you this life eternal". (John 5:39, LB)

The first crisis to hit the young church was an argument over the literal authority of the Old Testament. Dozens of scriptures in the Old Testament stated clearly and unequivocally that God expected all males to be circumcised. Yet Christian preachers like Paul and Barnabus did not feel that they should lay this burden on non-Jews who wanted to become Christians. But the fundamentalists among them insisted that "the Gentiles must be circumcised and required to obey the law of Moses" (Acts 15:5, NIV). After long debate, the delegates to this first church

council meeting voted unanimously *not* to continue to promote this commandment from the Old Testament. This vote had the full approval of the Holy Spirit, if we are to believe the Bible on this account (Acts, chapter 15).

If the Bible is literally and absolutely true, then it is literally and absolutely true that the church, not single, isolated individuals, but the whole church, has the authority, indeed the responsibility, to determine what parts of the Old Testament have special relevance to the contemporary situation and what parts of the Old Testament should be discarded.

The Science of the Creationists

Creationism is by no means a uniform, monolithic kind of position. There is considerable diversity among those who call themselves creationists. For example, when it comes to estimating the age of the earth, some early Biblical scholars estimated that the earth was about 6,000 years old. Bishop James Ussher dated creation in the year 4004 B.C. Melanchthon, a contemporary of Luther, dated it in 3963 B.C. John Lightfoot, Vice-chancelor of the University of Cambridge, said that man was created by the Trinity on October 23, 4004 B.C., at nine o'clock in the morning. (24, i, p.9) Most modern creationists hold to a date about 10,000 years ago, apparently in direct contradiction to the arithmetic of the Bible. Kenneth Taylor maintains that "the Bible permits millions of years as easily as thousands, and is not helpful in deciding this question as to when Adam was created". (21, p.31)

Part of the problem of dating the creation revolves around the concept of the "days" of the creation narrative. Some creationists insist on a literal, 24 hour day. A Jehovah's Witness publication says, "each creative day was at least 7,000 years in length". (3, p.7) Taylor says again, " . . . each day in Genesis was millions of years duration. . . . Man is perhaps up to a million years old. So we are talking about days 100,000,000 years long." (21, p. 49) And Harry Rimmer asks, "Are the Days of Genesis literal days of twenty four hours each, or are they periods of time? To that question we can only reply, 'We do not know' and then set forth the evidence that shows also why we *cannot know*". (19, p.12)

So, take your choice!

One of the points that creationists have emphasized in their discussions is the fact that in rocks older than the Cambrian Period we do not find any fossils. They would date the beginning of creation with the Cambrian Period, but with a much shortened time scale. But in the last three or four decades we have found fossils in pre-Cambrian rocks, some as old as 3.5 billion years. (6, p. 41-50) Hundreds of fossils of microorganisms of many sorts, both prokaryotic and eukaryotic, have been found. In spite of that, creationists have maintained until just recently that this is not true. John N. Moore said in 1970 that " . . . not a single indisputable fossil prior to the Cambrian Period has been found". (15, p.6) And Gish said almost the same thing in 1972, " . . . not a single indisputable fossil has ever been found in the Precambrian rocks". (23, p. 728) However, by 1978 Gish was willing to say, "Not a single, indisputable metazoan fossil has ever been found in Precambrian rocks". (10, p.62)

Original Creation of Species

Another observation on the quality of the creationists' science has to do with their concept of the "species" and just what really was created. Due chiefly to the work of Linnaeus, each species of plant and animal was conceived as created originally in its present form and had persisted without change from the beginning. (8, p.295) As Darwin put it in an essay, " . . . until recently the great majority

of naturalists believed that species were immutable productions and had been separately created". (4, p.v)

This might have been easy to believe when there were only a few thousand species. Linnaeus described some over 4,000 species himself during his lifetime and at the end of his taxonomic career he was beginning to raise questions as to whether all of those species had been created in that exact form. But he didn't dare speak out on that subject. (18) But with hundreds of thousands of species, or a million species, and some say as many as ten million species of plants and animals, it becomes very difficult to conceive of all of these species having been created at one time and persisting unchanged since that time.

Creationists gradually conceded that the original creation was some level of higher category, above the species level, which then produced variations, even closely related species. Morris said that " . . . the creation model . . . postulates that all the *basic kinds* of plants and animals were specially created and did not evolve from other kinds at all . . . the term kind is used to denote the originally created entity, within which variation could take place . . . the 'family' (in the Linnaean sense) is a good approximation [of the basic kind]." (17, p.29, emphasis added)

So, what was created according to Morris, was a series of primordial families that eventually diversified to form a variety of genera and countless species. If this is true, then we might consider that all of the orchids, belonging to the family Orchidaceae (about 15,000 species) are all descended from the first original orchid "kind" created in the beginning. This sounds very much like "descent with modification" (Darwin's words).

Gish follows this same use of the "basic kind" as the original created entity, including "all the variants which have been derived from a single stock". (10, p.34) He says that mammals represent a "basic kind". This is an entire class. But then he says that lemurs, monkeys, apes and men are the "basic kinds" (these are families). In the very next sentence he insists that gibbons, orangutans, chimpanzees and gorillas are each different basic kinds (these are genera). Among the invertebrates Gish lists protozoa, sponges, jellyfish, worms, snails, trilobites, lobsters and bees as "different kinds". Remember, these are the originally created entities that have produced a great variety of modern descendents, all "derived from a single stock" (Gish's own words).

Take worms, for example. The original primordial worm ancestor has produced such a variety of modern descendents that zoologists have classified them into 16 different phylums, including Mesozoa, Platyhelminthes, Rhyncocoela, Nematoda, Nematomorpha, Rotifera, Gastrotricha, Kinorhyncha, Acanthocephala, Annelida, Echiurida, Sipunculida, Priapuloidea, Phoronida, Pogonophora, and Hemichordata. This includes the development of acoelomate, pseudocoelomate and eucoelomate descendents from the original "kind" of worm. There is also included in this geneological sequence both protostomate and deuterostomate forms.

Either Gish did not mean what he said, or he doesn't know what he is talking about. The kindest thing one can say here is that Gish displays extensive unfamiliarity with invertebrate taxonomy.

Creationism as a Science

I have presented creationism as an outgrowth of a minority religious viewpoint rather than a scientific hypothesis. Even the most avid promoters admit that creationism is not a scientific study.

Taylor writes that " . . . the creationist [does not come] to his conclusion

from evidence acceptable under the usual laws of scientific proof". (21, p.10) And Gish says in his book, "We do not know how the Creator created, what processes he used, for He used processes which are not now operating anywhere in the natural universe . . . We cannot discover by scientific investigations anything about the creative processes used by the Creator". (10, p.40)

Henry Morris, the head of the Institute for Creation Research, and perhaps the chief spokesman for the creationism movement, says, "Creation cannot be proved. Creation is not now taking place, so far as can be observed. Therefore, it was accomplished sometime in the past, *if at all*, and thus is inaccessible to the scientific method. It is impossible to devise a scientific experiment to describe the creative process, or even to ascertain whether such a process can take place. The Creator does not create at the whim of a scientist". (16, p.5, emphasis added)

Yet these writers refer to their proposition as "scientific creationism", and this impresses lots of people. They ask state legislators and judges to mandate that creationism should be included in science courses in our schools. And legislators and judges often fear to do otherwise.

Many people appear confused and uncertain because we refer to the "theory" of evolution. To the average person, a theory denotes something very tentative, not grounded in fact, not proved. Most biologists would regard evolution as an established fact, while the various theories are proposed explanations of how the process took place.

President Reagan, during the campaign in 1980, fell into this trap when he said, "Well, it is a theory, a scientific theory only, and it has in recent years been challenged in the world of science and is not yet believed in the scientific community to be as infallible as it once was believed". (6, p.28) He no doubt picked up a lot of votes when he made this statement before a fundamentalist audience in Dallas, Texas.

Creationists talk often of a "creation model". But most of the creationist literature consists of arguments against and criticism of the data, evidence and interpretation of data presented in evolution literature. If one removes the anti-evolution portions from the creationists' books, there isn't much left.

A model in this sense would be a description, as far as is known, of the events and sequences of events during and since the creation. It is what one might have observed and recorded with a hidden camera. There never has been much of a creation model developed along these lines.

Creationists delight to use words which attribute the most outlandish and ridiculous positions or expressions to their opponents, the evolutionists. Thus, a fundamentalist preacher in Maryland said recently, "The Lord is our Creator. He is our maker, and we should pay attention to what he says. He knows we need to operate. Someone came to me and said, 'I heard you believe God created the world in seven days, each twenty four hours long. Is that right?' I said, 'No, that is not true. I believe he did it in six twenty-four hour days, and rested on the seventh. I believe in a God who has power.' The Bible clearly teaches that God made the heaven and the earth in six days. The morning and the evening were the first day, and when he came down to the seventh day, he rested. The liberals believe in a powerless God, who cannot do anything. They say he can't save a soul, can't heal anybody, can't create a world, can't take you to heaven, and that he doesn't even have a heaven. Their God doesn't have any power. It takes him a billion years just to get a frog. Then another million years to get a monkey, and another million to get a liberal, and they still have the monkey. These pseudo-scientists sit around and try to explain how, through some kind of houdinism, why a head of cabbage didn't develop a brain like a man's head.

That is the kind of stuff they come up with. They would say, well, a head of cabbage ought to have as much smarts as a man. The sad thing is, they try to pawn this off on our little boys and girls. You have to be a stooge to believe that. You need special training". (7)

Even Henry Morris challenges evolutionists to discover " . . . what is the directing program that instructs a population of worms to develop themselves into a population of, say, crocodiles?" (17, p.21)

There are no scientific arguments against creationism because it is a religious belief, held by those whose primary faith commitment is the acceptance of the Bible as the verbally inspired, inerrant, direct Word of God.

There can be no religious arguments against evolution because science is based on observable, repeatable and testable empirical data. It does not depend on revelation.

Revelation is always current and contemporary; it is always new and exciting. When we attempt to memorialize the revelation, we inhibit further spiritual growth and development and force other persons into an arbitrary and sterile relationship to that revelation. In the so-called "transfiguration experience" of Peter, James and John on the mountaintop, the disciples wanted to create three monuments, one to Moses, one to Elijah, and one to Jesus, honoring Him as one of the three great leaders of their people. A voice seemed to say to the disciples: don't do it! "This is my beloved son; listen to him". (Mark 9:2-7) Don't build these monuments. Jesus is with you now; listen to him now!

In the play, Inherit the Wind, which dramatized the famous Scopes trial in Tennessee in 1925, the two attorneys, Brady and Drummond (representing William Jennings Bryan and Clarence Darrow) meet in a private, intimate setting. "We were good friends once", said Brady. "I was always glad for your support. What happened between us? There used to be a mutuality of understanding and admiration. Why is it, my old friend, that you have moved so far away from me?" Drummond replies slowly, "All motion is relative. Perhaps it is you who have moved away—by standing still." (12, p.60)

We can never go back!

We can never go back before Darwin, when nobody ever heard of natural selection, and gene mutation, and ecology. We can never go back to the geology before Lyell, or the physics before Newton, or the astronomy before Copernicus.

We cannot stand still, either in our religion or our science. We can only try to find new ways of living out our love for God and our fellowmen. We can only continue to learn more about our world and the way it works.

We can never go back!

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HIGHLIGHTS IN HERPETOLOGY: IT'S NOT ALL SNAKES

SHERMAN A. MINTON, M.D.
Indiana University School of Medicine,
Indianapolis, Indiana 46202

When Father Schmelz contacted me regarding a title for this address, I must have been mildly annoyed with someone who thought that herpetology is only the study of snakes. I suppose I should have been grateful that anyone knows that much about what a herpetologist is and does. Long ago when I was beginning my study of the amphibians and reptiles of Indiana, I saw the name of a physician who had been practicing in New Albany about the turn of the century listed as collector of a rather unusual snake in a museum collection. He had died about the time I was born, but New Albany is my home town, and I wrote to one of his relatives, a nice, middle-aged lady, saying that I didn't know her uncle had been a herpetologist and did she know what had happened to his collections? She replied that the doctor knew a lot about herbs because he made up many of his own medicines, and maybe I should be more careful with my spelling.

What I want to share with you are some stories from the lives of amphibians and reptiles that have intrigued me and I hope will be of interest to other members of the Academy. The first of these begins in 1827 when Jacob Green described a rather ordinary looking bluish-grey salamander from the campus of Jefferson College in Pennsylvania giving it the name *Salamandra jeffersoniana* or Jefferson's salamander. For the next hundred years, nothing very exciting happened concerning this salamander. It proved to be one of the commoner salamanders of the northeastern United States and ranged north in eastern Canada about as far as a salamander can survive. Like many wide-ranging animals, it showed considerable geographic variation. Some workers split it into several species, while others lumped it into one. It was removed from the genus *Salamandra* and put in the genus *Ambystoma*, and the general features of its life history were learned. Jefferson's salamander is almost unique among northeastern salamanders in being a winter breeder. Almost any time after the first of the year that there are two or three days of temperatures in the 40s or 50s accompanied by rain or heavy snow melt, these salamanders will emerge from their burrows and move to shallow ponds in woods or at the edge of woods. In the ponds, males and females find each other, and there is a courtship ritual in which the two rub and nuzzle each other until the male deposits a conical spermatophore on a twig or leaf, and the female pinches off the cap of sperm between the lips of her vent. Males may deposit as many as twenty spermatophores, and each female will pick up perhaps a half dozen, so there are usually a good many left over. In most years, the salamanders remain a month or so in the ponds and later are joined by two or three related species which have a similar breeding pattern, although the courtship rituals differ. After fertilization, females lay clumps of eggs encased in jelly. These hatch as the water warms, and the larval salamanders transform into adults about the end of May or early June in the midwest.

The first person to notice that there was something unusual about the life history of Jefferson's salamander was Wesley Clanton, a University of Michigan zoologist, who was studying the species near Ann Arbor about 1930. He found the population here consisted of "two confusingly similar forms" that he designated as dark and light. Moreover, the light form consisted entirely of females. These females mated with dark males to produce more females. In some sites this led

to a highly unbalanced sex ratio that Clanton felt would result in extinction of both forms. However he did not follow up his observations, and things rested there for almost twenty years. About 1950 I came briefly into the picture because I was working on the herpetology of Indiana. At that time I knew Jefferson's salamander from the populations around New Albany where they looked very much like the animal Green described from Pennsylvania and from the Indiana Dunes and Jasper-Pulaski State Forest where they were smaller and darker with prominent blue-white flecks. Both populations had a reasonable proportion of males to females. As I accumulated more material, I found intermediate individuals, nearly all of them females from north of Indianapolis. Eventually I concluded that the dark, blue-flecked northern form was a distinct species for which I revived the old name *Ambystoma laterale*. I speculated that hybridization between *jeffersonianum* and *laterale* produced only females, a situation that seemed to hold little future for the hybrid populations, since a growing number of females would be competing for an ever decreasing number of males.

The next really big advance came about 1960 when another University of Michigan scientist, Thomas Uzzell, did a great deal of field and laboratory work on these salamanders and came up with some interesting conclusions. One was that the all-female populations had a triploid number of chromosomes instead of the diploid number normal in sexually reproducing organisms. Also these triploids seemed to be of two types, one, to which he applied the name *platineum*, was similar to *Ambystoma jeffersonianum* and the other, given the name *tremlayi*, was very similar to *A. laterale*. He found that the mode of reproduction in these triploids is gynogenesis in which sperm are necessary to initiate development of the eggs but contribute nothing genetically. He reasoned that the sperm would probably come from one or the other of the related diploid species, since males of unrelated species would probably not court the triploid females nor would the females be stimulated to pick up spermatophores of males of unrelated species.

Since Uzzell's work in the 60s the problem has become more complicated. As Uzzell and I collected in Indiana ponds where triploid salamanders were found, very rarely did we find males of either related diploid species, but we always found good numbers of males of some ambystomid salamander, usually the smallmouth salamander, *Ambystoma texanum*. This is a rather nondescript salamander not too different in appearance from animals of the *jeffersonianum* complex. But in some other ponds the triploids were found only with the spotted salamander (*Abmystoma maculatum*) which is quite different in appearance and courtship behavior. And, although it has not been proved, it seems very likely that the triploids are using sperm of unrelated ambystomid salamanders to initiate development of their eggs. If this is not the case, then true parthenogenesis must be occurring. More recently there have been reports of diploid eggs being found in egg masses laid by triploid salamanders and of true hybrids between salamanders of the *jeffersonianum* complex and the smallmouth salamander which may itself be a composite of two species. It looks like there is material here for midwestern zoologists to work on for quite some time.

Going from small woodland amphibians to very large marine reptiles, I want next to say something of the wanderings of sea turtles. There are seven species of sea turtles. All are large and have breeding ranges in the tropics or subtropics. They are the most commercially valuable of reptiles and are variously utilized for meat, eggs, shell, leather, and oil. There are entire island and maritime communities whose culture and economy is as closely linked to the sea turtle as ours is to the automobile. And, particularly in the last century, sea turtles have been badly over exploited. Only comparatively recently have we become aware of this,

and efforts to reverse the trend are just beginning to show some success. In order to protect the turtles, it has been necessary to learn a great deal about their biology, particularly their movements to and from nesting grounds. Adult turtles are fairly easy to mark, usually by putting a metal tag on the front flipper, and thousands have been tagged at nesting grounds. Through the recovery of tagged turtles, we are slowly learning something of their wanderings.

As they leave the water to nest, female turtles leave their signature on sandy beaches throughout the tropics, although not in the numbers they did a century or two ago. There are two main stages to nesting, first construction of a large, shallow body pit by strong sweeps with the fore flippers and second the digging of the small, deep egg pit with the hind flippers. Once the second stage has begun, nothing short of dynamite will deter the female from her task. After nesting, the female returns to the sea where she often mates with males that have followed her to the nesting beach. A female may lay two or three clutches of eggs in a season, but it appears that no species nests annually. Most seem to be on a two or three year cycle with the possibility of a four year cycle for some. Several species of turtles may use the same nesting area.

Most of the information we have about sea turtles is based on the green turtle, *Chelonia mydas*; however, the broad pattern seems similar for all species. Although turtles of all species show great fidelity to nesting grounds, only the green seems to have well defined feeding grounds as well. This may be because it is the only species that is wholly an herbivore as an adult. The migration between feeding and nesting grounds may be long or fairly short. One of the longest and most interesting migrations is made between feeding grounds along the bulge of Brazil and nesting grounds at Ascension, a tiny island about half way between South America and Africa. An Air Force song of World War II had a line that went, "If you miss Ascension, your wife gets a pension." indicating that even with modern navigational equipment the island wasn't the easiest place to find. But the turtles find it, and they make the trip out and back without eating. Moreover, the baby turtles somehow find their way back to the coasts of South America where they mature. Other green turtles that feed along the Brazilian bulge make a long migration along the north coast of South America to nest on Surinam beaches. All studies indicate these turtles never nest at Ascension, nor do Ascension turtles ever nest at Surinam. The well studied green turtle population at Tortuguero on the Atlantic side of Costa Rica migrates for the most part only a short distance to feed along the Mosquito Coast of Nicaragua, but occasional marked turtles from this population have been found throughout the Caribbean.

The leatherback (*Dermochelys coriacea*) is the largest of sea turtles, indeed the largest of living reptiles if only weight is considered; record specimens may reach about 500 kg. Its shell is not a massive bony structure but rather a mosaic of many small bones embedded in a thick skin. The form is beautifully streamlined with seven low ridges on the upper side and five below. The leatherback also has the widest range of any reptile. Although most of its breeding sites are within the tropics, there are about 200 sightings or captures of leatherbacks in northern European waters from Iceland to well above the Arctic Circle off the north tip of Norway. There are some 88 records for New England and Canadian waters. In the north Pacific, records go from the Gulf of Alaska to the northernmost islands of Japan. There are fewer records from the far southern seas, but this may only mean there are fewer observers there. Data from tagged leatherbacks indicate that their dispersal from nesting beaches seems to be random. The most remote spots from home base that turtles tagged at Surinam beaches

have been found are the coasts of Ghana and New Jersey. In each case, the trip took about a year.

In the Atlantic, loggerheads (*Caretta caretta*) are recorded about as far north as leatherbacks, but there are far fewer sightings. I am not so sure now as I was a few years ago that these leatherbacks and loggerheads sighted in far northern or southern seas often make it back to the tropics. The leatherback has a mechanism, not altogether understood, for keeping its core temperature as much as 17°C. above that of the sea. It is also known that a water temperature of about 11°C. is very likely to be lethal to these turtles. So if they get into northern waters, they had better get back before the temperature falls to this critical point. We have no idea how many do. A recent report of activities of leatherbacks and loggerheads off the coast of Florida as observed from a spotter plane indicated that the turtles in August seemed to be avoiding the Gulf Stream as though they did not want to be carried north by its current.

We still have no good idea how sea turtles navigate. Another and probably related puzzle is the so-called lost year of the baby turtle. The baby turtles, 80 to 100 to a nest, literally erupt from the sand about sixty days after the eggs are laid. Hundreds are destroyed by crabs and other predators before they ever reach the water, and doubtless many others are eaten soon after they enter the sea. But there comes a time soon after the young turtle gets to the sea that it just literally disappears. There are almost no specimens of small turtles from hatchling size to the size that we estimate from growth of captives to be that of a yearling, and most of those that do exist were taken from stomachs of predators such as sharks. However, what we think happens is that baby turtles swim very vigorously away from the shore until they are picked up by a current, and probably presence of favorable currents is one thing that determines if a beach will be a good one for turtle nesting. Then they are carried passively by the current for a great distance. As the baby turtle drifts, its brain is storing countless bits of chemical and physical data about each current and perhaps surprisingly unique to it. When the time comes for the turtle to return to the waters of its birth, it calls on this store of information to guide it back. Turtles also have a sun compass sense as many cold-blooded vertebrates do, and there is some evidence they may orient with the earth's magnetic field. But we still have a great deal to learn about sea turtle navigation, and about sea turtles generally.

Finally I want to talk about lizards and snakes, and more specifically about some novel ways these reptiles use their tails. Many vertebrates use the tail as an aid in locomotion, in swimming, climbing, flying, jumping and running; lizards and snakes use it for locomotion, too, but they may do other things as well. Anyone who has tried to catch small lizards learns that they readily sacrifice their tail to a pursuer. The broken tail vigorously writhes and twists, distracting the hunter and often allowing the lizard to escape. The tail is rather promptly regenerated, the new tail generally being less perfect than the original. Sometimes a forked tail or one with several branches is produced.

In some other lizards, the tail is heavy, firmly attached, and spiny, making it an uncomfortable object for a potential predator to grasp. It is curious the way this adaptation crops up sporadically in unrelated groups of lizards. It is seen among the sungazers (*Cordylus*) a family endemic to southern Africa, the mastigures (*Uromastix*) of the Afro-Asian desert belt that are members of widespread agamid family, the spiny-tailed iguanas (*Enyaliosaurus*) of Mexico, again a member of a widespread family, and in a few Australian species of the cosmopolitan skink family.

Another use of the tail in defense is that of the varanids or monitors. These

large Old World lizards use their tail actively as a whiplash. A monitor about a meter long can give a stinging blow, and larger ones have been reported to disable dogs in this manner.

A very strange tail defense is used by a few small Australian geckos of the genus *Diplodactylus*. If they are injured or severely threatened, they eject from their tail strands of a material rather like spider silk. First reports indicated the material came from the prominent rows of papillae on the tail; actually it comes from cracks between the papillae. Although this is obviously a defensive tactic, it is by no means clear how the material affects a possible enemy. One group of scientists put some of the material on mealworms and offered them to chicks. The chicks didn't seem to like the worms as well as untreated ones, but there was no evidence the substance was toxic or even highly distasteful. I smelled the secretion and put a bit on my tongue. It had a faint odor rather like crushed maple leaves, and there was no taste at all. I have but one rather wild speculation as to how and against what it may be used. These lizards live under the bark of standing trees. The bark must be loose enough that the lizards can move about but not so loose that they are vulnerable to snakes, birds and other predators. The same habitat is used by large flat spiders the Australians call "huntsmen". These spiders could probably kill even the adult geckos and could certainly eat the young ones. In the cramped quarters of their habitat I can imagine these silky strands entangling or impeding a spider enough that the gecko might escape.

Lizards may engage in threat displays that involve use of the tail. In the case of the fat-tail gecko (*Eublepharis macularius*) of southwest Asia, the lizard rises high on its legs, arches its back, and waves its tail about rather like a scorpion preparing to sting. It may also open its mouth and make an odd noise something between a squeal and a hiss. In Pakistan this gecko, known as "hun-khun", was the most feared of reptiles. Its bite or contact with any of its body fluids were said to be instantly fatal.

In our western deserts probably the speediest lizards are species of the genera *Callisaurus* and *Holbrookia*. They dart almost too fast for the eye to follow across the flat, bare soil, stop suddenly and flatten their body, becoming practically invisible. And then, for no apparent reason, they will often raise the tail, showing the conspicuously banded black-and-white undersurface and wave it slowly about almost as though they were inviting attention. Then they will streak off again usually in a different direction. In Baluchistan I saw a toad-headed lizard (*Phrynocephalus maculatus*) do something rather similar. I found these lizards only in an area that was flat and incredibly barren—there was literally no place for a lizard to hide. They usually sat on rocks, and as you approached them they would curl the tail in a spiral and slowly lash it back and forth. Then they would run and flatten against the soil, but they did not then elevate the tail as the American lizards do.

Tail displays are common among snakes. Several species of coral snakes when disturbed will thrash about with the tail elevated and the tip curled on itself so that it looks somewhat like the head. Another venomous snake, *Maticora bivirgata* of southeast Asia, does much the same thing. In this species, both head and tail are bright orange and the body blue-black. Western subspecies of the ringneck snake (*Diadophis punctatus*) when alarmed often go into elaborate contortions in which the tail is curled into a tight spiral showing the undersurface which is bright red in contrast to the yellow or orange belly.

Using the tail for sound production reaches its acme in the rattlesnakes. They seem to be taking special advantage of an old and widespread serpent habit

of vibrating the tip of the tail when alarmed. In a suitable medium such as dry leaves this can make a distinctive sound. I once thought that this trait in such nonvenomous snakes as rat snakes and king snakes was an attempt to imitate the rattlesnake, however it shows up in many parts of the world where there are no rattlesnakes to imitate.

Curiously enough, in the deserts of southwest Asia is a lizard that uses its tail for sound production. It is a medium-size gecko (*Teratoscincus scincus*), and it makes a sound by undulating its tail in such a way that the large plates on its dorsal surface rasp against each other. The sound is a very faint one, and I missed it on my first encounter with this strange reptile. But it can be heard under the right conditions. Again, I can only speculate what benefit it is to the lizard to make this sound. In the great sand dunes of Baluchistan, *Teratoscincus* lives with a deadly little snake, the saw-scaled viper, that has its own curious method of sound production. It inflates its body with air and rubs loops together so that the serrate scales make a sizzling sound. The sound the lizard makes is similar but much fainter, however in a burrow or underground cavity it might be enough to frighten an enemy.

Another function of the tail is fat storage. This is seen most frequently in lizards that live in desert areas and exist on a feast or famine regimen. The gila monster of our Southwest is a good example. The closely related Mexican beaded lizard does not have such a fat tail, apparently because it lives in a more mesic region. The uncommon little gecko *Teratolepis* of the Sind Desert in Pakistan is another species that stores fat in its tail as does the wood gecko (*Diplodactylus vittatus*) of eastern Australia. Tails of these fat-tail geckos usually break only at the base, so the small predator that seizes one gets a fair meal for its effort. The regenerated tail often becomes more bulbous than the original. The shingleback (*Trachydosaurus rugosus*) is a large, slow, armored Australian skink whose stumpy tail can be almost as large as its head, and it really is hard to tell at a glance which end is which. This could serve to misdirect the attack of an enemy. Some small sand boas of the genus *Eryx* may use the thick, blunt tail to divert an attacker. I saw this illustrated quite graphically one night when collecting in the Sind desert. A sand boa about 70 cm long lay in a tight coil, its tail chewed raw and bleeding. Tracks showed it had been attacked by a fox or small jackal. Had the animal seized its head, the snake would almost certainly have been killed; as it was, it suffered only trivial injury.

I saw a West Indian dwarf boa (*Tropidophis*) use this defense when threatened, making a compact coil that hid its head but left the tail exposed. This snake does not have a particularly blunt tail, but the tip is pale orange and constrasts strongly with the body color. I suspect that this species may use its tail in another way that is well known among snakes but has not been described for lizards, namely as a lure for prey. Most of the snakes that are known or suspected to use tail-luring are venomous species that strike from ambush and feed on lizards, frogs, and perhaps small birds. The conspicuous tail tip presumably resembles a grub or other insect larva. In most species, only young snakes use tail-luring. Raymond Ditmars was one of the first to describe how new-born copperheads wiggled the yellow tips of their tails when small frogs were put in their cage. The cantil, a Central American relative of the copperhead, holds its tail upright to make the light tip more conspicuous. The late Ross Allen described how frogs were attracted by the lure and even sometimes bit it before the snake caught them. Toads were also attracted, but the snakes would not attack them. In the dangerous Central American pit viper *Bothrops asper* (one of several species known as fer-de-lance) only the young males have yellow tail tips, and luring has not been observed

Young males of the Saharan sand viper (*Cerastes vipera*) have yellow tail tips and females black; the significance of this is unknown.

The heavy-bodied Australian death adder belongs to the elapid or cobra family but looks for all the world like a viper. It too has a tail lure, in fact it has the most elaborate tail lure I have seen. The tip of the tail is slightly widened and flattened, the scales are arranged to simulate segments, and the color is yellow with a few light and dark flecks. The resemblance to an insect larva is surprising. For several months I had a death adder in a glass cage near my desk. When the snake was hungry, it raised its tail displaying the lure. I could not determine if it attracted prey; if I dropped a lizard in the cage, the snake seized and swallowed it at once. As I watched this snake, I wondered if perhaps a similar tail lure was not once on a pit viper with the habit of tail vibrating. For one reason or another, sound production served the species better than luring prey, and, given a few million years of selection pressures, the rattle of the rattlesnakes evolved.

Returning to lizards, tail shape, size, and color may be important in social interactions. Recent work indicates that individuals of the little side-blotch lizard (*Uta stansburniana*) of our western states lose their place in the social hierarchy if they lose a third or more of their tail. Another study indicates that the bright blue tails characteristic of many young skinks of North America and eastern Asia inhibit aggressive behavior by adult males of their own species. To confuse things, there are young skinks in Australia, Asia, and the western United States that have red or orange tails, and a little sandburrowing species in Florida has a tail that may be blue, orange, brown or violet. In other lizard families such as the anoles there are tail crests and other ornaments that probably help in defending territory and selecting mates. Lizard ethology is a whole different game with the players frisking about in sunny spots on six continents. I recommend them for your study and enjoyment.

ANTHROPOLOGY

Chairman: Curtis H. Tomak
Indiana State Highway Commission
2370 Cherry Drive, Martinsville, Indiana 46151

Chairman-Elect: Gary D. Ellis
Indiana Division of Historic Preservation
Indianapolis, Indiana 46201

ABSTRACTS

Riverton Lithic Technology at the Wint Site, Bartholomew County, Indiana. C. Michael Anslinger, Indiana State University, Terre Haute, Indiana 47809.—— A technological and functional analysis of the chipped-stone tools and debitage from the Wint Site (12-B-95) indicates selective chert procurement and utilization patterns during the Late Archaic. Also, two distinct reduction technologies were identified. Implications for Riverton settlement system analysis, and future research goals are discussed.

A Preliminary Report on Recent Explorations at Fox Island, Allen County, Indiana. Diane E. Beynon, Department of Anthropology, Indiana University-Purdue University at Fort Wayne, Fort Wayne, Indiana 46805, and Paul Jean Provost, Department of Anthropology. Indiana University-Purdue University at Fort Wayne, Fort Wayne, Indiana 46805.——This paper will be a preliminary report on the archaeological excavations recently completed at Fox Island County Park in Allen County. The excavation was carried out by the authors as part of the annual Field School in Archaeology being run by the Department of Anthropology at Indiana University-Purdue University at Fort Wayne.

12-Wb-90, Lithic Source Areas and Settlement Pattern Analysis in the Lower Mississinewa Drainage. Donald R. Cochran and William Wepler, Ball State University, Muncie, Indiana 47306.——A controlled surface collection from 12-Wb-90, an eroded site on the shore of the Mississinewa Reservoir, produced an unusual number of endscrapers and exotic chert artifacts when compared with the other 110 sites recorded during a 1981 survey of the reservoir. An analysis of the artifacts and the site location and comparison with other sites in the area revealed that recorded lithic source area sites do not fit the usual characterization of quarry or workshop sites. The location of chert outcrops in the Lower Mississinewa drainage allowed for selection of site locations that were in proximity to chert and other resources so that separate scheduling of quarry and workshop activity was unnecessary.

Experimental Archaeology—The Building of a Controlled Prehistoric Site on a 50 Meter by 50 Meter Area. Phil McClure and Robert E. Pace, Wabash Valley Archaeological Society.——Activities such as house construction, food processing and preparation, pit construction, tool making, pottery making and firing.

The methods used are conducted in aboriginal manor within the woodland time frame.

This site will be constructed in order to determine time and physical energy expenditures required for habitation of the area.

Five to ten years later excavations will be conducted, and the data used to evaluate the methods of present day recovery procedures.

Some Regional Variations in the Late Archaic. ROBERT E. PACE, Indiana State University, Terre Haute, Indiana 47809.—— Test excavations at the Bluegrass Site (W-162) in 1981-82 have established a close association with the French Lick Phase of the Late Archaic. Other Late Archaic sites in southwest Indiana are of the French Lick Phase. Differences have been observed between contemporary Late Archaic sites in southeast Indiana, leading to a suggestion of a "Falls Phase." Differences have also been observed between these two and contemporary sites in areas of east central Indiana. It is suggested that the Howell Site (Franklin County), tested in 1979, is representative of a third regional phase, the "Howell Phase." Similarities and differences between these three expressions of the Late Archaic are discussed.

Albee Component at the Cooke Site (P-5) Parke County, Late Woodland in the Central Wabash Valley. ROBERT E. PACE AND DANIEL P. THIEL, Indiana State University, Terre Haute, Indiana 47809.—— Albee Culture is represented as one of the multi-components at the Cooke Site (12-P-5) in Parke County. Carbon-14 dates place it between 1100-1300 A.D. Sub-plowzone features have produced evidence of corn as an important item in the diet. Ceramics suggest the site is closely related to a Late Woodland complex represented by a series of sites in west-central Indiana, with regional associations in central Indiana and the Illinois River Valley.

Mechanical Stress in a Late Woodland Population. MELANIE A. RESEIGH, Muncie, Indiana 47302.—— The Commissary Site, near New Castle, Indiana, was excavated by the Ball State Archaeological Field School during the 1966-71 seasons. This paper deals only with those adults excavated that were complete enough for an analysis of mechanical stress of the bones. During this analysis it was noted that the cervical vertebrae of some individuals, along with other skeletal areas, had indications of heavy use. This paper will discuss possible cultural causes for the stresses involved.

A Survey of the Upland Forest and Prairie Conjunction in Vermillion County, Indiana. WILLIAM E. RESEIGH, Ball State University, Muncie, Indiana, 47306.—— Surveys of upland areas are only now becoming common, and have begun to change our perceptions of prehistoric patterns of land use. A survey of the Newport Army Ammunition Plant, which contains areas of dissected and undissected upland prairie, and river flood plain, is compared with information from other prairie and upland studies.

The results show that site frequency in prehistoric times is very similar for prairie and undissected upland, and that there are possibly concentrations of sites in areas of dissected upland which are close to both undissected upland and prairie areas.

Bono: A Late Archaic Shell Midden in Lawrence County, Indiana. CURTIS H. TOMAK, Indiana Department of Highways, Indianapolis, Indiana 46204.—— The Bono Site occupies about two acres of upland overlooking bottomland of the East Fork of White River in Lawrence County. The author initially surveyed the site in 1980 and in 1982 conducted an archaeological excavation there which was funded by the Indiana Academy of Science.

The site possesses large midden areas of dark soil mixed with a large quantity of mussel shell and non-midden areas of lighter soil containing little shell. Excavation was done in both types of area. Midden was found to extend below

plowzone, and some archaeological material was recovered from beneath the midden. In addition, one or two aboriginal features were found.

A considerable amount of archaeological debris besides shell is present at Bono. This material occurs in the midden and in the non-midden areas but with apparent differences in content between the two types of area. Included among the material recovered from the site are points, bifaces, drills, scrapers, axes, campstones, bone tools, broken rock, chert debris, animal bone, and charred plant remains. The artifacts indicate that Bono was occupied mainly during the later half of the Archaic period.

The Melvin Site—12-B-401. MARK WOLFAL, Columbus, Indiana, 47201.——The Melvin Site is located in the southern portion of Bartholomew County on the first terrace of a flood plain.

The site was tested in early March of 1981 by the Wabash Valley Archaeological Society. The test consisted of seven 2 meter x 2 meter squares and two 1 meter x 1 meter squares. The test squares were selected by the location of surface material scattered over a large area of the terrace.

Pottery sherds collected from the surface and during the test excavation included curvilinear guilloch, cord impressed, collard rims, and smooth.

The pot sherds, along with the other material, show the site to be a single component site; and very closely related to the Bowen Site located in Marion County, thus associated with the Oliver Phase.

The single date available for the Oliver Phase sites is 890 AD ± 100 years. The single date for the Melvin Site is 955 AD ± 90 years.

A Preliminary Archaeological Survey of Boone County, Indiana

Douglas R. Mitchell
Glendale, Arizona 85308

Introduction

Beginning in the late summer of 1980 and continuing through the spring of 1982, the author conducted a surface survey of parts of central and northwestern Boone County, Indiana. The survey was done largely as a one man—weekend operation. The purpose of the survey was to locate prehistoric sites in an area in which relatively little archaeological work had been done. While a rigorous research and survey design was not utilized for this project, it is felt that the data gathered will be helpful in forming the basis for future research in this area.

Environmental Setting

Boone County is located in central Indiana. It is within the Scottsburg Lowland and Bluffton Upland bedrock physiographic units. These consist primarily of Devonian and Mississippian bedrock formations which contain shales, siltstones, sandstone, limestone, and dolomite. The bedrock is covered by till of the Trafalgar formation deposited during the Wisconsin glaciation. The topography of the county is characterized by a flat to gently rolling glacial plain, known as the Tipton Till Plain. (4) This broad, flat plain is modified slightly by drainageways, with Sugar Creek being the most prominent in the county.

Boone County lies within two major drainage basins, the Wabash and the White. The northwestern part of the county is in the Raccoon subdivision of the Wabash basin. The south-central portion is in the Eel subdivision and the southeastern portion is in the West Fork subdivision of the White River basin. Sugar Creek, with its headwaters in southwestern Tipton and southeastern Clinton Counties, flows southwest emptying into the Wabash River in Parke County. Prairie Creek drains part of central Boone County and flows northwesterly emptying into Sugar Creek near Thorntown.

Subsequent to the recession of the glaciers from central Indiana and the attendant climatic change, the cool, moist climate allowed the formation of a coniferous forest. This coniferous forest was followed, about 6000 B.C., by the slow development of three major types of vegetation; deciduous woodlands, prairie, and wet prairie or swamps. (6) Boone County appears to have had some of each of these distinct habitats.

According to the General Land Office (GLO) Surveys done in the county in the 1820s, (1) the forested areas were characterized by Beech, Sugar Maple, Oak, Ash, Hickory, some Walnut and Elm. Mention is also frequently made of prairie and swamps. Prior to European habitation of this area and the consequent land clearing and draining, the varied vegetational zones would have supported an abundant and diverse faunal population. Mumford estimated that as of 1816 there were 66 species of mammals in Indiana. (2) Fish, reptiles, amphibians, and fowl would have also been available for aboriginal consumption.

Survey and Results

The survey procedure consisted of examining areas of high site potential such as rises and ridges near water sources. The areas focused on were portions

of the Sugar and Prairie Creek stream valleys. Most of the survey was conducted in upland areas, with only a limited amount of survey done in the floodplain.

A total of 206 sites were found in Boone County and 10 sites in Montgomery County. Ninety of these sites produced material diagnostic of one or more cultrual periods, for a total of 116 diagnostic components.

One site, possibly two, produced evidence of Paleo Indian occupation. Site 12Bo116 is a multicomponent site located on a remnant terrace near Prairie Creek. One broken fluted point base was found on this site. Several endscrapers were also found but may belong to a later component at this site. Site 12Bo190, located on an upland bluff north of Sugar Creek, produced a point base which is possibly Paleo Indian. The unnotched point base is basally thinned but lacks the characteristic basal and lateral grinding.

The Early Archaic Period is represented by 28 components. Points of this period found during the survey include Kirk, Thebes, Charleston corner notched and the Bifurcate base types. The Bifurcate base types (LeCroy, Kanawha, etc.) seem to be the most common. All but two of the sites with Early Archaic components were found in upland settings.

The Middle Archaic Period is ill-defined in Central Indiana and no points of this period were identified. The Late Archaic Period however, is well represented. Fifty-eight components were found evidenced by a variety of side notched and stemmed points. Nutting stones, one full grooved ax, and a bannerstone fragment were found on sites of this period. Sites are located in both upland and floodplain settings during this time, Riverton points (7) were located on eight sites, all in upland settings (Figure 1).

FIGURE 1. *Early Archaic points, top row. Late Archaic points, middle and bottom rows.*

The Terminal Archaic/Early Woodland Period is represented by six components. Three of the points found are round base Adena points and nearly all of the TA/EW points found appear to be made from Harrison County chert. All sites with TA/EW components were found in an upland setting. Four Snyders-like points and two Lowe Flared Base-like points were found representing the Middle Woodland and Late Middle Woodland Periods, respectively. One Snyders-like point was found in an upland setting.

The Late Woodland Period is represented more frequently than the preceding Woodland Periods. Sixteen components were found most commonly evidenced by triangular points. These sites were found in both upland and floodplain settings (Figure 2). Four celts and one pendant or gorget fragment were also found which are indicative of the Woodland Period. No prehistoric pottery was found on any of the sites and there is no evidence for Mississippian occupation of this area.

FIGURE 2. *Terminal Archaic/Early Woodland points, top row. Middle Woodland points, middle row. Late Middle Woodland point, far left. Late Woodland points, bottom row.*

There was a historic Indian village in and around Thorntown occupied by the Eel River tribe of the Miamis during the early 1800s; however, no evidence of this occupation was found during the survey. No early historic structures were located although scattered historic debris such as glass, ceramics, and nails was not uncommon in fields. This probably represents debris from late 19th century—early 20th century farmsteads.

A variety of lithic resources are available throughout the county in the glacial till. Hardstone cobbles, used for hammer, grinding and nutting stones, are common. Slate can also be found in the glacial gravels. Small cobbles of chert are

also common and account for a good portion of the chert tools and debris found on sites. Chert of nonlocal origin was also found on sites in the form of finished tools and debris, with Attica or Independence chert being the most common. Small quantities of Harrison County chert, Harrodsburg chert, (5) Upper Mercer chert, Burlington chert, and Cataract chert (3) also occur. A new chert source was located by the survey and is discussed below.

In the northwestern portion of the county an area of bedrock is exposed along the banks of Sugar Creek. The bedrock exposed here is primarily shale of Mississippian Age. Beginning just east of the Montgomery/Boone County line, a lense of chert is exposed and extends for about 600-700 feet to the east. Above and below the chert is siltstone and shale. Beginning approximately 500 feet east of the county line and extending for 150-200 feet to the east, a second lense of chert is exposed which is three to four feet higher than the previously mentioned lense.

The lower chert lense, here referred to as Sugar Creek Chert, is a tabular type. The lense is 9 to 12 inches thick and large blocks 2 to 3 feet long can be seen eroding down into the creek. In color it is bluish gray to olive gray variegated with white cloudy streaks. It is not uncommon to find pieces containing quartz crystal inclusions. This chert weathers to a light brown and may also weather to a purplish-reddish gray although this latter color may be a result of heat treatment. The chert is often fine-grained and fairly homogenous but may contain numerous fracture planes.

The chert lense above it, exposed for a shorter distance, is similar in texture but is tan to chocolate brown in color. It will be referred to as Sugar Creek chert—tan variety. This source does not appear to have utilized as extensively as the gray chert which may be a result of its limited accessibility.

Several large workshop sites were found in Boone and Montgomery Counties near the Sugar Creek chert source. The heaviest concentration of workshop sites occurs within approximately a one mile radius of the chert source. This chert is easily accessible from Sugar Creek and quarrying activities probably involved gathering chert which had eroded onto the stream bank or prying chunks out of the exposed chert lense. The material from these sites typically consists of chert chunks and debris, broken bifaces in various stages of manufacture, and occasional hammerstones. One of the largest sites, 12Bo39, is located on a bluff directly south of the chert out-crop. This site covers an area of nearly three acres. Limited, uncontrolled surface collections from 12Bo39 have produced over 20 bifaces and biface fragments (Figure 3), many pieces of chert debris and large chert chunks, hammerstones, and some projectile points and tools. Fire cracked stone is also scattered over the site.

One Late Woodland triangular point fragment, all of the bifaces and all of the debris are of Sugar Creek chert. The points and tools are made of a variety of cherts and indicate hunting and food processing activities. If these artifacts are contemporary with the workshop activities, then their presence on this and other workshop sites suggests that small aboriginal groups were living at these sites while processing chert.

Some of the chert debris is purplish-gray which may be a result of heat treatment or weathering, as previously mentioned. Experiments by the author show that heating this chert in an oven will produce a purplish-reddish tint. It is not known however, how this affects the knapping qualities of the chert.

There is a relative lack of finished tools made from Sugar Creek chert at 12Bo39 and other workshop sites. This suggests that the chert was worked into blanks or preforms at or near the source for transport. Material surface collected

FIGURE 3. *Bifaces and hammerstone from Site 12Bo39.*

FIGURE 4. *Bifaces from probable cache at Site 12Bo98.*

from a probable plowed-up cache at site 12Bo98 would tend to support this idea. Located approximately 16 miles southeast of the source, 12Bo98 produced 25 rough bifaces and 16 chert chunks, all of Sugar Creek chert (Figure 4). Five Late Archaic side-notched points found on this site are also of Sugar Creek chert. The bifaces were probably made near the chert source and, along with some additional unworked raw material, transported to this site. Whether the bifaces were meant to be further modified or used as heavy tools is not known. The lack of Sugar Creek chert debris at 12Bo98 confirms that they were not made or modified at the site.

Sugar Creek chert was available and utilized throughout prehistory. Points made out of this chert are evident during the Paleo Indian, Early Archaic, Late Archaic, Middle Woodland, and Late Woodland Cultural Periods. In addition, cutting and scraping tools were frequently made of this chert.

The spatial distribution of this chert is not well known. Sugar Creek chert artifacts and debris have been seen by the author in Boone, Montgomery, Clinton, Hamilton, Tippecanoe, and Hendricks Counties. Curtis Tomak (pers comm.) recently reported two Early Archaic points from Greene County which appear to be made from Sugar Creek chert. Further survey work and analysis of existing collections will provide more information on the distribution of this chert.

The survey was preliminary in the sense that additional survey work, further analysis of the existing collections, interviews with local artifact collectors, and analysis of inter- and intra-site distributions would undoubtedly produce a more complete picture. From the data recovered, however, it can be seen that utilization of small, upland tributary resource zones occurred during the Paleo Indian Period, moreso during the Early Archaic Period, but was most intense during the Late Archaic Period. Little evidence is seen for the Early and Middle Woodland Periods, but increasing use is seen again during the Late Woodland Period. Further work in Boone County and other parts of Central Indiana will aid in understanding the prehistory of this area.

Acknowledgments

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"Blowing" Family Savings Zapotec Style: A Theoretical Dilemma

Carl O'Nell
Department of Anthropology, University of Notre Dame, Notre Dame, Indiana 46556
and
Nancy O'Nell
Purdue University, North Central, Westville, Indiana 46391

The Zapotec Indians, with whom we did our research, live in southern Mexico chiefly in the state of Oaxaca. They are identified by their speaking any one of several Zapotecan dialects as a primary language and by living a traditional way of life that finds its roots in a pre-Columbian past. Although essentially a rural people at present, they are the descendants of peoples who evolved one of the earliest and most notable of meso-american civilizations comprised of city-states about 400 B.C. and terminating in an empire around 700 A.D. (5) (7).

While basically subsistence farmers many Zapotecs produce goods exceeding family needs. Such surpluses and goods resulting from specialized crafts and husbandry have been exchanged in regional markets since at least early colonial times (2). Monetary resources from such exchanges have not been used for capital investment, and only sparingly for family needs and personal wants. Rather large amounts of accumulated monetary resources traditionally have been consumed through ritual expenditures perceived as contributing to community benefit. The principal vehicle for directing funds to community benefit has been the custom of participation in the *mayordomia.*

Briefly, the *mayordomia* is a widespread custom in mesoamerican peasantry systems which calls for the individual sponsorship of religious feasts. A man may volunteer for or be named to the position of sponsor. In either case the sponsor, called a *mayordomo,* takes on the burden of financing a religious feast for the benefit of his community. The economic burden is called a *cargo* in Spanish, and for this reason anthropologists often refer to the whole as the *cargo* system (6).

The cultural roots of the *cargo* system antedate the conquest of the New World by the Spanish, relfecting indigenous customs of ritual sponorship economically costly to individual sponsors. Utilizing the customs they already found in operation, the Spanish conquerors added the dimension of prestige to expenditures made in the service of the saints. As one writer puts it, the *cargo* " . . . system takes from those who have, in order to make all men have nots. By liquidating the surpluses, it makes all men rich in sacred experience but poor in earthly goods."(8)

Another writer, who sees the *cargo* system as just one type of customary behavior reducing inequalities in wealth between people in various primitive and peasant societies, refers to these systems as "leveling mechanism" (3). Analytically the leveling mechanism emphasizes the importance of individual contribution to community welfare and service while it deemphasizes the importance of individual economic success. Individual economic sacrifices are compensated for by community conferred prestige.

For hundreds of years and indeed up to the present Zapotec farmers and their families have been willing "to blow" family savings by participating in the *mayordomia.* Heads of families have been willing to take on the *cargos* seen as so important by themselves, their neighbors and fellow townsmen, and in return to receive honor and prestige for the financial sacrifices so made. The system

has been notably resistant to change and because it contributes to the maintenance of equilibrium in the total society its persistence contributes to stability in Zapotec society generally.

The ethnographic evidence is that the *cargo* system may finally be beginning to give way to a system favoring economic competition, the accumulation of wealth, personal discretion in the use of monetary resources, and the purchase of goods for personal and family pleasure and profit. There is a real question however about how long it might take a capital forming economic system to overtake and supplant the *cargo* system. The cultural roots of the *cargo* system run deep into the soil of a social order brought about by the fusion of indigenous Indian systems and a Spanish economic system which had not yet emerged into the industrial age.

It cannot simply be assumed that the *cargo* system will give way to some more modern system of a use of accumulated savings at the family level. For the most part the shift from spending household savings from community service to family use is an effect and not a cause of the breakdown of the *cargo* system. In places where the *cargo* system no longer remains a vital element in one's participation in community affairs we often are able to observe that it has been displaced functionally by other elements which serve to maintain a balance of equilibrium in community social dynamics. It is tempting to think of the *cargo* system only as a siphoning agent for individual and family resources. The chief quality of the cargo system lies in the fact that it serves as an important factor in the maintenance of social equilibrium at the community level.

One of the more fruitful formulations explaining the eventual demise of the *cargo* system comes from Frank Cancian (1). Cancian develops his hypothesis about the passing of the *cargo* system from his excellent functional analysis of the *mayordomia* in the elaborate *cargo* system in the Mayan community of Zinacantan.

Briefly, Cancian maintains that currently increasing population and wealth in Zinacantan stand as threats to the functionality of the *cargo* system in the maintenance of equilibrium and stability in the community. The equilibrating system which previously functioned in precisely that way is now overburdened by a steadily increasing population of adult males who have the economic resources and the desire to take on *cargos* but who cannot be accommodated by positions traditionally open to them. Increasingly resources which would be consumed in *cargo* expenditures are being diverted to the purchase of goods becoming attractive to would be participants and their families. Cancian refers to many of these goods as "*ladino* symbols" because they are taking on values beyond their utilitarian ends and because ownership of such goods often has prestige value among non-Indians or *ladinos*. *Ladino* symbols include thing which reflect wealth and social status for non-Indians—special housing, cars, radios, bicycles, westernized clothing, and increasingly more important, education beyond the primary level. Bit by bit prestige of a type is beginning to accrue to the ownership and use of *ladino* symbols by Indians, although this prestige is not comparable at present to that gained through participation in the traditional *cargo* system.

Says Cancian, "The integration of Zinacantan society which has been a consequence of the *cargo* system will inevitably break down". (1) His analysis disposes him to predict that as *cargos* become less of an economic burden (through greater potential for wealth for individuals), while other economic outlets become more accessible and important to people, the more the *cargo* system will be threatened. In consequence, the prestige which comes from filling *cargos* will decline while prestige accruing to the ownership of *ladino* symbols will increase, gradually to replace that which derives from participation in the *cargo* system. The end result

will be complete erosion of the *cargo* system which will no longer serve economic leveling and social prestige functions.

Fortunately in our field work we have been able to observe the results of the demise of the *cargo* system in a Zapotec community, called Escalerita. Our observations span a period of about fifteen years but our informant data allows us to reconstruct conditions when the *cargo* system was intact, and to analyze those conditions which led to the collapse of the system.

In many respects Escalerita is similar to Zinacantan. Both are "cultural communities" in the sense that residents have a decided community orientation to the world, perceiving themselves in community-centric terms relative to people in neighboring communities. People in both communities have traditional approaches to life and regard their own way of life as having as much validity as that of the non-Indian. Two important variables distinguish the communities. Zinacantan is large and for several decades has been relatively prosperous. Escalerita is small and for much of this century has been relatively impoverished.

Prior to 1950, when the *cargo* system was officially dissolved in Escalerita, there were four levels of *cargos*. The system could accommodate from 8 to 15 yearly *mayordomias* although it seems quite certain that not all positions were filled every year. Men were involved in *cargos* from ages 15 through 55 years. Positions in the *mayordomia* were matched with positions in the civil political hierarchy so that assuming a cargo in the religious structure permitted a man the possibility of assuming a *cargo* of commensurate rank in the political structure. A man's prestige in the system was reflected not only in the kinds and levels of *cargos* he could assume in the religious structure but also in the ranking of the positions open to him in the political structure. All men entered the system at the lower levels, hoping through participation to achieve positions at the higher levels. No man, for example, was considered fit for the position of town president unless he had served in five *cargos* in the *mayordomia* and had occupied certain lesser political offices. A man could not occupy the most prestigious position of *alcalde* (mayor) unless he had served as town president and in the two major *mayordomias* in the highest grade of the *cargo* system.

Any man could volunteer for a *mayordomia* at a level appropriate to his age and prior service. But a committee also existed to name men to serve if no one volunteered for the essential positions. Official lists were kept recording *cargo* services.

Evidence is that the system functioned well in the early decades of this century. Probably because of the Revolution (1910-1917) conditions changed which affected the *cargo* system in Escalerita. Political *cargos* declined in importance, some becoming non-existent. This loosened the all important connections between religious and political positions which apparently undermined the prestige of the *mayordomia*. Economic decline accompanied political decline and *cargos* became unbearable economic burdens for many people.

Fewer and fewer people controlled the resources necessary for participating in the *mayordomia*. With fewer volunteers the council acted more frequently in naming men to unfilled important, and of course more expensive, *cargos*. Enforced participation and outright refusals became the rule rather than the exception. Prestige through service under these circumstances became questionable, particularly when service in the *mayordomia* was not matched by opportunities to serve in the political structure which traditionally validated prestige gained in the *mayordomia*. The small size of the community exaccerbated the generally unfavorable conditions.

The president of Excalerita in 1950 with considerable, but by no means

unanimous, support dissolved the *mayordomia*. His act erased the obligatory aspect of the system and gave official recognition to the fact that large numbers of people were simply no longer able to participate in the system (4). Enough resistance to the act resulted so that for at least a few years it appeared that the *mayordomia* might be reinstated as a compulsory system of *cargos*. And the custom of the *mayordomia* did not disappear immediately. Even into the early 1970's some men still periodically volunteered to support one or the other of the major feasts. Although they thought of themselved as *mayordomos* taking on *cargos*, they rarely assumed full expenses for the feasts as was customary in the past. Initially most townspeople saw the expenses as "unnecessary." Now there is an almost unanimous tendency to see such expenditures as "foolish" or "wastefuls" on the part of the individuals who might undertake them.

Ecomonic recovery occurred after the dissolution of the *mayordomia*. But it is interesting, and important, to note that no major effort has been made to reinstitute the *cargo* system as a result of recovery. With economic recovery a number of community-wide enterprises have been undertaken which would not have been successful had it not been for assessments leveled on heads of households by the town authorities. Some of these assessments were made proportional to the estimated wealth of the individuals assessed conforming to the spirit of the *cargo* system. As a result the community now enjoys a complete six year primary school, an all weather road, a deep well with a pump to supply clean drinking water, and electricity for both public and private use. Such assessments of funds for the public good do not normally exhaust family savings however.

What is apparent in the thirty year period since the *mayordomia* was abolished is the observable fact that people are not "blowing" family savings in the support of *cargos* but in the purchase and use of what Cancian calls "*ladino* symbols." For example, in 1950 there were no radios in the community; in 1965 there were 5 or 6; in 1971 there were about 20. By 1981 there were at least twice as many and one person had already purchased a television set. Prior to 1968 no one owned a vehicle of any sort in Escalerita. In that year the first bicycle and the first truck were purchased. By 1981, many bicycles, several trucks and one farm tractor had been purchased. The quantity of tiems purchased is not as important to note as is the qualitative shift from ritual expenditures to personal expenditures, reflecting it would seem the complete and final disappearance of the *cargo* system in this Zapotec town.

Chiefly of interest to us is how we might best explain the cultural factors involved in the complicated processes we have observed. Over the years we have witnessed the demise of the *cargo* system in Escalerita, but under conditions which appear to be opposite those posited by Cancian for Zinacantan. Because our observations appear to be at variance with those of Cancian in many respects it might seem that his formulation has little utility for us in understanding the case familiar to us. A problem arises from trying to explain contradictory cases with a theoretical formulation that analytically applies well only to one. We in fact however believe that Cancian's formulation offers potential for the generation of some more general theoretical construct to help us . . . and others . . . understand (and perhaps predict) the collapse of the *cargo* system wherever found, facilitating its replacement by other economic mechanisms. Currently we are working with some more broadly based theoretical constructs to see which of them can best accommodate an understanding of these contradictory cases thereby helping us to address the dilemma we have but briefly outlined in this paper.

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BOTANY

Chairman: KATHRYN WILSON
Department of Biology, Indiana University-Purdue University
at Indianapolis, Indianapolis, Indiana 46223

Chairman—Elect: L. DAVID POLLEY
Department of Biology, Wabash College, Crawfordsville, Indiana 47933

ABSTRACTS

Growth of Green Ash Seedlings Influenced by Available Soil Phosphorus but not Mycorrhizal Inoculation. D. S. EGEL and P. E. POPE, Department of Forestry and Natural Resources, Purdue University, West Lafayette, Indiana 47907.— Vesicular arbuscular (VA) mycorrhizae are reported to improve the survival and enhance the growth of herbaceous and certain woody plant species on droughty, nutrient deficient sites. A high probability for survival and rapid early growth of outplanted tree seedlings are paramount for a successful reforestation program. This study reports the influence of the mycorrhizal fungus *Glomus macrocarpum* (Tul. and Tul.) on the growth, and nitrogen and phosphorus uptake of green ash (*Fraxinus pennsylvanica*, Marsh.) seedlings grown over a wide range of available soil P Levels.

Green ash seedlings, grown in a glasshouse for 21 weeks, were harvested at 3 week intervals. Seedlings were grown at 10, 30, 70, 110 and 125 ppm Bray I extractable soil phosphorus and were either inoculated or not with *Glomus macrocarpum* at the rate of 380 spores/kg of autoclaved loamy sand soil mix. At each harvest, seedling height, diameter, fresh and dry weight by component (root and stem plus foliage) were measured and 10-20% of roots (fresh weight basis) were taken for determination of the percentage of mycorrhizal colonization. Percentage foliar phosphorus and nitrogen were also measured for each seedling.

Neither seedling growth nor percentage foliar phosphorus was affected by mycorrhizal inoculation but both increased with increasing soil phosphorus levels. Percentage mycorrhizal colonization decreased with increasing soil phosphorus levels and tended to decrease with time. Shoot/root ratio of seedlings was unaffected by either mycorrhizal inoculation or soil phosphorus levels.

Abnormal Dependence on Potassium in *Chlamydomonas*: Evidence for Transport Deficient Mutants. C. JAMES FRANCOEUR and L. DAVID POLLEY, Wabash College, Crawfordsville, Indiana 47933—— Mutants of *Chlamydomonas reinhardii* which exhibit a dependence abnormally high concentrations of potassium for growth have been isolated. These have been characterized genetically and physiologically. Here we report the results of our studies in which the kinetics of potassium (^{86}Rb) transport were examined.

Potassium transport in wild type strain R3 + NM conforms to simple Michaelis-Menten kinetics. When these cells are grown on medium supplemented with 10mMK, the V_{max} of ^{86}Rb uptake is 25 nmoles Rb/hr x 10^6 cells and the K_n is 0.75 mM. When these cells are stressed for potassium by a 24 hour starvation in medium lacking potassium, both the affinity for potassium and the capacity of transport increase. By contrast, two mutant strains which exhibit nearly identical kinetic parameters when grown in 10mMK can not absorb any potassium after 24 hours of starvation. The third mutant strain exhibits slightly increased

capacity when grown in 10mMK, but there is no change in kinetic parameters when stressed by starvation. The relationship between these observations and possible mechanisms which regulate potassium transport will be discussed.

Genetic and Morphological Differences in Twelve Populations of *Collinsia verna* (Scrophulariaccae). JUDITH K. GREENLEE, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.——*Collinsia verna* (Scrophulariaceae), a spring flowering annual, occurs in small isolated populations. The focus of the study was to determine population dynamics and genetics of *C. verna*, its relationship to other species in the genus, and the status of *C. verna*, in Indiana.

Populations were collected from Indiana, bordering Kentucky counties and southern Illinois. Plants were collected in the budding stage and crosses were made in all possible combinations between populations. Morphological and cytological data obtained show differences between populations based on gross external characters as well as chiasmata frequency and meiotic configurations.

Electrophoretic techniques being employed show polymorphisms which are diagnostic between *Collinsia* species. Between populations, within populations and F1 crosses are currently being analyzed. Microspectrophotometric techniques being employed show differences in the amount of DNA between and within species and populations.

C. verna populations were found to be decreasing and disappearing from sites disturbed by development of parks, mowing, home building, and trash dumping.

Micromorphology and Chemistry of Fossil Laticifers from Brown Coal Deposits of the Eocene. PAUL G. MAHLBERG and DONALD FIELDS, Department of Biology, Indiana University, Bloomington, Indiana 47405.——Cellular remains from brown coal deposits of the Geiseltal, near Halle, were determined to be a fossil form of the nonarticulated laticifer. A cellulosic wall was often present surrounding the cells which retained a tubular shape during their preservation. Small holes, possibly artifactual in nature, occurred at irregular intervals along the cells. The cells contained a dense, yet spong-like, content primarily polyisoprenoid in composition. Rapid polymerization of the rubber in these cells undoubtedly aided in the preservation of these cells. These contents, which filled the entire cell lumen, occasionally included structures that may have represented cellular organelles or nuclei in the cell. Branches were present along laticifer axes and, as for branched nonarticulated laticifers in extant plants, these branches projected in different directions from the principal axis of this cell. Gas-liquid chromatography of acetone extracts derived from cellular contents, demonstrated the presence of several sterols which formed a characteristic sterol profile for these remains. The cellular morphology and sterol profile of these laticifers can be employed as markers, in comparative studies with extant plants, to identify the species origin of these laticifers.

The Effect of Organic Chelators and Divalent Cations on Nickel Toxicity to Two Species of Green Algae. LORI H. NICHOLS and DAVID F. SPENCER. Department of Biology, Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana 46223——Cultures of two algal species (*Ankistrodesmus falcatus* var. *acicularis* and *Scenedesmus quadricauda*) were maintained in a chemically defined medium. Total nickel concentration was 10.2 micromolar, a concentration previously shown to be toxic to these species. Organic chelators, ethylenediamine tetraacetic acid (EDTA) and nitrilotriacetic acid (NTA), were used to adjust the concentration of free divalent nickel. Algal response was measured by growth

rate and cell size. These parameters were inversely related to the concentration of free divalent nickel and independent of the total nickel concentration in the medium. Equilibrium calculations for the medium suggest that changes in growth were due to changes in the concentration of free divalent nickel and not changes in the availability of other nutrients and trace metals. The effect of CA, Mg, and Mn on nickel toxicity was also examined. Results indicated that within the ranges tested, Ca, Mg, and Mn did not reverse nickel toxicity for *Ankistrodesmus falcatus* var. *acicularis*.

A Fairy Ring of *Chlorophyllum molybdites* (Fr.) Sacc. PHILIP A. ORPURT, Department of Biology, Manchester College, North Manchester, Indiana 46962. —— The occurrence of a sizeable ring of large mushrooms in the yard of a nearby farm was called to the attention of the author in September 1981. In response to a request that the fungus be identified to determine its edibility, the site was visited where photographs and specimens were obtained.

Chlorophyllum molybdites (*Lepiota morgani* Peck) is known to form large fairy rings throughout our area, however, the particular occurrence of this very often impressive mushroom was the most spectacular I had ever observed. The ring of 78 large sporophores measured ten feet in diameter. The largest specimen was 10½ inches tall with a cap 8 inches broad. Many of the other individuals were not much smaller. While some of the sporophores possessed gills which were essentially white, the more mature specimens were beginning to exhibit an olivaceous coloration. Spore prints soon revealed that the spores were indeed an olive color which made possible a positive identification.

Specimen site: The Robert Bechtold farm, T29N, R7E, Section 2, Wabash Co., Indiana. Date of collection: September 7, 1981. Specimens are on deposit at Manchester College and at Southern Illinois University.

Pisolithus tinctorius Colonization and Site Conditions Influence Establishment of Red Oak Seedlings. P. E. POPE, Department of Forestry and Natural Resources, Purdue University, West Layfayette, Indiana 47907. —— Planting oak seedlings as a method of reforestation has been only moderately successful. Once planted, the seedlings characteristically exhibit little above ground height or diameter growth of 3-5 years. The ectomycorrhizal forming fungus (*Pisolithus tictorius*) which successfully colonizes and enhances the outplanting survival and growth of southern pines also colonizes red oak.

Seedling nursery beds of red oak (*Quercus rubra*) were inoculated (2.5 $1/m^2$) with sterilized or viable vegetative fungal inoculum of *Pisolithus tinctorius* and seedlings grown for one year. Dormant barefoot seedlings were planted in replicated 25 tree plots in March 1978 on three major soil types to evaluate the influence of soil-site conditions and mycorrhizal inoculation on seedling survival, development and nutrient uptake. After 3 years, seedling survival ranged from 81 to 93% and was significantly influenced by soil-site and initial mycorrhizal condition. Annual and 3 year growth increments (cm^3) were greater for inoculated seedlings but the interaction with the soil-site component was also significant. For all sites, foliar P concentrations were greater for seedlings inoculated with the test fungus but foliar P levels did not differ significantly among sites for the inoculated seedlings. For the inoculated seedlings, the percentage of feeder roots infected by the test fungus declined over time on all sites but after 3 years had not reached the natural infection levels of the non-inoculated seedlings.

Dissolution and Chemical Composition of Laticifer Starch in *Euphorbia pulcherrima*. STEVEN R. SPILATRO and PAUL G. MAHLBERG, Department of Biology, Indiana

University, Bloomington, Indiana 47405.—This investigation was undertaken in order to better understand the slow rate of starch degradation when laticifer starch granules are treated with hydrolytic enzymes. GLC of trimethylsilylated derivatives of acid hydrolized laticifer starch revealed glucose to be the only carbohydrate present. 'Blue value' and iodine binding capacity determinations showed laticifer starch to contain 30-35% amylose. Suspension of laticifer starch granules in DMSO at 100°C or lower resulted in partial (<65%) dissolution after 40 hours. Dispersion at 125°C resulted in 90% dissolution of the starch granules after 9 hours and complete dissolution within 16 hours. The refractory nature of laticifer starch granules to enzymatic hydrolysis appeared not to be related to the presence of any unusual carbohydrates within the granule. The amylose content of the granule, although higher than the typical 20-25% found in seed and tuber sources, also does not indicate a barrier to enzymic hydrolysis. The unusually slow rate of dissolution of the starch granules, however, does indicate that the molecular association of the starch molecules may be sufficiently strong to produce a structure inaccessible to hydrolytic enzymes.

Kinetin and Abscisic Acid Increase the Rate of Water Permeability across Lipid Bilayers. WILLIAM STILLWELL and PAUL HESTER, Department of Biology, Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana 46223.—Kinetin and abscisic acid are shown to greatly enhance the rate of water permeability across lipid bilayer membranes. Rapid water permeability is followed by measuring spectrophotometrically the rate of liposome swelling in hypotonic solution or the rate of liposome shrinking in hypertonic solution as a function of kinetin or abscisic acid included in the bathing solution. The kinetin effect is measured from 16.3 μM to 2.7 mM, from pH 2.0 to 11.0, at temperatures from 10°C to 50°C, with membranes made from both natural (egg lecithin and asolectin) and synthetic phosphatidylcholines. The effect is greatest for membranes in the more fluid, liquid crystalline state. Abscisic acid produces a much greater increase in water permeability with membranes composed of natural phospholipid mixtures as opposed to synthetic phosphatidylcholines. The abscisic acid effect is dependent on the presence of phosphatidylethanolamine in the membrane. These experiments indicate that these plant growth substances may exert their initial, rapid time effect on the lipid bilayer portion of membranes.

Callus Growth and Embryogenesis in Tissue Cultures of *Asclepias tuberosa* L. (Asclepiadaceae). KATHRYN J. WILSON, Department of Biology, Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana 46223.—Tissue cultures of *Asclepias* species provide an experimental system suitable for specifically defining the biochemical and physical parameters of differentiation and growth of the cell type, the laticifer. Since laticifers typically present in *Asclepias* members differentiate in zygotic heart stage embryos, experiments were undertaken to determine culture media supporting rapid callus growth and ultimately promoting the most prolific asexual embryogenesis. Stem explants callused most rapidly when derived from seedlings no older than 25-30 days. To derive callus, explants were maintained on several combinations of 2,4-D(0-4.0 mg/l) and benzyladenine (BA) (0-2.5 mg/l) incorporated into a basic Murashige-Skoog (MS) medium. After 33 days greatest callus growth was obtained on media supplemented with 0.5 mg/l 2,4-D and 0.5 mg/l BA. Significantly greater numbers of embryoids were obtained on media supplemented with 0.5 mg/l 2,4-D combined with 1.0, 2.0, or 2.5 mg/l BA and their numbers were increased by transferring one month old callus to MS medium supplemented with 0.5 mg/l BA and either no or a relatively high concentration of (6.0 mg/l) of 2,4-D. High concentrations of auxin promote embryoid

development most similar to normal embryogeny. The effect on laticifer differentiation in embryoids proliferated on high auxin concentrations currently is being investigated.

A Compilation of Plant Diseases and Disorders In Indiana—1982

GAIL E. RUHL, RICHARD X. LATIN, PAUL C. PECKNOLD and DONALD H. SCOTT
Department of Botany and Plant Pathology
Purdue University, West Lafayette, Indiana 47907

Introduction

The Plant Diagnostic Clinic (PDC) in the Department of Botany and Plant Pathology at Purdue University is a service of the Cooperative Extension Service, Purdue Agricultural Experiment Station. The clinic provides free service for interested persons through the county extension system, for accurate identification of weeds, plant diseases and plant disorders. This paper is a compilation of plant diseases and disorders which were diagnosed in the clinic from January 1 through November 15, 1982. Such a compilation is an invaluable tool in determining the problem areas in which extension personnel need to concentrate. Ultimately, comparisons of yearly disease/disorder compilations could provide additional insight for epidemiological studies in Indiana.

Methods

Plant specimens are submitted to the PDC from county extension agents, homeowners, growers, nursery operators, consultants, and others. Specimens are diagnosed visually or by culturing the pathogen on selected media. Some virus diseases are diagnosed by the ELISA technique or by the leaf dip technique utilizing the electron microscope. Once a disease or disorder is diagnosed, appropriate control measures are suggested. A computerized log system is used to summarize data for epidemiological studies, to provide readily accessible reference materials on samples and for report generation. A summary of the samples diagnosed from January 1 through November 15, 1982 is given in Table 1.

Results

As in previous years (1,2) weather and site related problems were common in Indiana during the 1982 growing season (Table 1). An extremely hard winter, highlighted by record breaking low temperatures in early January and February caused severe cold injury problems throughout the state. Early dry spring weather precluded the development of many infectious diseases and allowed for optimal planting conditions. However, late spring rains enhanced the development of some foliar disease problems.

Overall, with good growth conditions and optimum dry down weather for warm season crops (corn and soybeans), disease losses were relatively light during 1982. However, some isolated infectious diseases did become widespread in drought and heat-stressed plants.

Tables 2-8 show the host plant, the disease or disorder diagnosed, the pathogen or cause of disorder, and the number of samples received.

Shade and Ornamental Trees

Diseases: Early dry spring weather precluded the development of shade tree anthracnose and primary scab on crabapples. However, late spring rain caused some leaf drop on sycamore due to anthracnose and secondary scab on crabapples. As in past years (1,2,3,4), *Diplodia* tip blight continued to be the primary disease

TABLE 1. *Plant Samples received in the Purdue Plant Diagnostic Clinic Jan. 1 through Nov. 18, 1982.*

Plant Specimen	Number of Samples	Disease	Disorder	Chemical	Nutritional
AGRONOMIC (18%)					
Corn	131	88	34	16	4
Soybeans	66	54	7	20	1
Small Grain	40	42	10	1	4
Forage Grasses and Legumes	36	32	4	0	6
Tobacco	1	0	1	0	0
ORNAMENTAL (43%)					
Trees-Shade and Ornamental	435	90	327	17	10
Shrubs and Groundcover	121	54	64	6	1
Flowers	52	38	11	2	1
House plants	31	6	21	1	3
FRUIT (8%)					
Tree Fruit	86	29	50	3	4
Small Fruit	35	24	17	2	0
VEGETABLE (12%)	178	81	52	16	19
TURFGRASS (2%)	34	18	13	1	2
PLANT IDENTIFICATION (13%)	190	–	–	–	–
FORWARDED TO ENTOMOLOGY (4%)	65	–	–	–	–
TOTAL	1501	556	611	85	55

[1]Problem caused by an infectious disease causing agent, e.g. fungus, bacterium,virus, mycoplasma, nematode.
[2]Problem caused by noninfectious environmental stress, e.g. wind, drought, heat, soil compaction.
[3]Problem caused by herbicide/pesticide misuse.
[4]Problem caused by a nutrient imbalance.

seen on Austrian, red and Scotch pines (Table 2). Occurrence of the recently discovered pine tree disease, pine wilt, caused by the pine wood nematode, *Bursaphelencus lignicolus*, did not appear to increase. *Verticillium* wilt of maple and redbud and Dutch elm disease were frequently reported during the summer. *Disorders*: Severe winter cold resulted in injury to a number of tree species; most noticeably affected were oriental cherry, redbud, dogwood and English oak. Extensive terminal twig death due to the severe cold was most severe in redbud. Pine, spruce, and hemlock showed extensive needle browning and twig death due to both cold and winter desiccation. The most frequently recorded disorders of maple, oak, and ash were scorch and dieback caused by a combination of stress factors including inadequate soil moisture. Leaf drop and decline were extensive on ash during mid-summer. The cause of this disorder on ash is unknown. Dieback of mountain ash caused by a combination of sun scald, canker, and borers, was frequently recorded. Iron chlorosis of oaks, especially pin oak, remains a problem in those areas of the state with a high soil pH.

Ornamentals

Diseases: As in past years (1,2,3), yew dieback was the most frequently recorded

TABLE 2. *Shade and Ornamental Trees—Diseases and Disorders*

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Abies (FIR)		
Tip Blight	*Diplodia pinea*	1
Miscellaneous Disorders		
Poor Vigor	Stress factors(s)	2
Winter Damage	Desiccation	1
Acer (MAPLE)		
Anthracnose	*Gloeosporium apocryptum*	2
Wilt	*Verticillium albo-atrum*	2
Canker	*Botrydiplodia* sp.	1
Leaf Spot	*Phyllosticta* sp.	2
Leaf Spot	Unidentified fungus	2
Heart Rot	Fungi/stress	1
Miscellaneous Disorders		
Scorch	Heat, wind, and drought	42
Dieback	Stress factor(s)	23
Herbicide Injury	Spray drift	8
Mechanical Injury	Hail	1
Chlorosis	Nutrient deficiency	2
Acer (BOX ELDER)		
Anthracnose	*Gnomonia* sp.	1
Leaf Spots	Unidentified fungus	1
Miscellaneous Disorders		
Dieback	Stress factor(s)	1
Betula (BIRCH)		
Leaf Spot	*Septoria* sp.	1
Miscellaneous Disorders		
Decline	Transplant shock	1
Scorch	Heat, wind, and drought	3
Carya (HICKORY)		
Miscellaneous Disorder		
Scorch	Heat, wind, and drought	3
Catalpa (CATALPA)		
Miscellaneous Disorder		
Scorch	Heat, wind, and drought	1
Celtis (HACKBERRY)		
Miscellaneous Disorder		
Decline	Stress factor(s)	1
Cercis (RED BUD)		
Wilt	*Verticillium albo-atrum*	2
Miscellaneous Disorders		
Decline	Improper pruning	1
Scorch	Heat, wind, and drought	4
Dieback	Stress factor(s)	1
Winter Injury	Desiccation	2
Cladastris (YELLOW WOOD)		
Miscellaneous Disorder		
Scorch	Heat, wind, and drought	1

TABLE 2.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Cornus (DOGWOOD)		
Leaf Spot	*Septoria* sp.	3
Miscellaneous Disorders		
Scorch	Heat, wind, and drought	8
Dieback	Stress factor(s)	2
Herbicide Injury	Spray drift	1
Decline	Unknown	2
Crataegus (HAWTHORN)		
Fire Blight	*Erwinia amylovora*	2
Miscellaneous Disorder		
Dieback	Stress factor(s)	2
Elaeagnus (RUSSIAN OLIVE)		
Canker	*Fusicoccum elaeagni*	4
Miscellaneous Disorder		
Decline	Stress factor(s)	2
Fagus (BEECH)		
Miscellaneous Disorders		
Scorch	Heat, wind, and drought	1
Decline	Stress factors(s)	1
Fraxinus (ASH)		
Petiole Anthracnose	*Gloeosporium aridum*	1
Miscellaneous Disorders		
Scorch	Heat, wind, and drought	8
Dieback	Stress factor(s)	6
Ilex (HOLLY TREE)		
Miscellaneous Disorder		
Chlorosis	Nutrient imbalance	1
Juglans (WALNUT)		
Downy Spot	*Microstroma* sp.	1
Leaf Spot	*Phyllosticta* sp.	1
Leaf Spot	Unidentified fungus	1
Miscellaneous Disorders		
Improper Fruit Set	Poor pollination	2
Scorch	Heat, wind, and drought	2
Herbicide Injury	Spray drift	1
Juniperus virginiana (RED CEDAR)		
Twig Blight	*Phomopsis juniperovora*	2
Miscellaneous Disorders		
Winter damage	Desiccation	3
Dieback	Stress factor(s)	2
Liriodendron (TULIP TREE)		
Powdery Mildew	*Erysiphe polygoni*	2
Leaf Spot	*Phyllosticta liriodenderi*	1
Miscellaneous Disorders		
Sooty mold	*Caphodium elongatum*	1
Scorch	Heat, wind, drought	5
Leaf Yellowing/Spotting	Natural stress	5
Chemical Injury	Spray drift	2
Decline	Stress factor(s)	2
Magnolia (MAGNOLIA)		
Miscellaneous Disorders		
Winter Damage	Desiccation	1
Scorch	Heat, wind, and drought	1
Malus (CRABAPPLE)		
Canker	Unidentified fungus	1
Firebright	*Erwinia amylovora*	1

TABLE 2.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Miscellaneous Disorders		
Winter Injury	Desiccation	2
Dieback	Stress factor(s)	2
Picea (SPRUCE)		
Canker	*Cytospora kunzei*	2
Needlecast	*Rhizosphaera kalkoffii*	1
Miscellaneous Disorders		
Winter Injury	Desiccation	7
Chemical Injury	Improper uses	2
Needle Tip Burn	Heat, wind, and drought	12
Mechanical Damage	Unidentified	2
Decline	Transplant shock	1
Cut Branches	Squirrel damage	1
Pinus (PINE)		
Tip Blight	*Diplodia pinea*	16
Needle Cast	*Lophodermium pinastri*	1
Needle Blight	*Dothistroma pini*	4
Pinewood Nematode	*Bursaphelenchus lignicolus*	2
Southern Fusiform Rust	*Cronartium fusiforme*	1
Root Rot	Unidentified fungus	1
Miscellaneous Disorders		
Canker	Animal damage	1
Decline	Poor drainage, site-stress	41
Winter Damage	Desiccation	12
Needle Tip Burn	Heat, wind, and drought	6
Sooty Mold	Insect honeydew secretions	6
Yellowing/Autumn (3rd yrs growth)	Natural	8
Mechanical Damage	Unidentified	1
Herbicide Injury	Spray drift	1
Platanus (SYCAMORE)		
Petiole Anthracnose	*Gnomonia veneta*	1
Powdery Mildew	*Oidium obductum*	1
Miscellaneous Disorder		
Scorch	Heat, wind, and drought	1
Populus (POPLAR, ASPEN, COTTONWOOD)		
Dieback	*Cytospora* sp.	1
Miscellaneous Disorder		
Decline	Site stress factor(s)	2
Twig Fall	Stress factor(s)	1
Scorch	Heat, wind, and drought	1
Prunus (PURPLE LEAF PLUM)		
Brown Rot	*Monilinia fructicola*	1
Miscellaneous Disorders		
Green Leaves	Genetic	1
Winter Damage	Desiccation	1
Herbicide Injury	Spray drift	1
Prunus (ORIENTAL CHERRY)		
Canker	*Cytospora* sp.	1
Miscellaneous Disorders		
Decline	Poor site	1
Winter Damage	Desiccation	5
Scorch	Stress factor(s)	1
Pyrus (ORNAMENTAL PEAR)		
Miscellaneous Disorder		
Scorch	Heat, wind, and drought	1

TABLE 2.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Quercus (OAK)		
Anthracnose	*Gnomonia quercina*	1
Miscellaneous Disorders		
Chlorosis (Pin Oak)	Iron deficiency	7
Dieback	Stress factor(s)	11
Scorch	Wind, heat, and drought	8
Chemical Injury	Spray drift	2
Decline	Transplant shock	1
Rhus (SUMAC)		
Wilt	*Verticillium* sp.	1
Miscellaneous Disorder		
Decline	Winter Injury	1
Robinia (LOCUST)		
Heart Rot	Wood rotting fungi	1
Miscellaneous Disorders		
Scorch	Heat, wind, and drought	3
Decline	Stress factor(s)	3
Salix (WILLOW)		
Leaf Spot	*Septogloeum* sp.	1
Canker	*Cytospora chrysosperma*	4
Miscellaneous Disorder		
Decline	Stress factor(s)	3
Sorbus (MOUNTAIN ASH)		
Fire Blight	*Erwinia amylovora*	1
Leaf Spot	*Septoria* sp.	3
Miscellaneous Disorders		
Canker	Sunscald/fungi complex	5
Scorch	Heat, wind, and drought	6
Dieback	Stress factor(s)	1
Tamarisk (TAMARIX)		
Miscellaneous Disorders		
Winter Damage	Desiccation	1
Scorch	Heat, wind, and drought	1
Decline	Stress factor(s)	2
Thuja (ARBOR VITAE)		
Nursery Blight	*Phomopsin* sp.	1
Miscellaneous Disorders		
Inner Needle Browning/Autumn	Natural	4
Winter Injury	Desiccation	1
Tilia (LINDEN)		
Miscellaneous Disorder		
Scorch	Heat, wind, and drought	2
Tsuga (HEMLOCK)		
Miscellaneous Disorder		
Winter Damage	Desiccation	6
Decline	Stress factor(s)	3
Ulmus (ELM)		
Dutch Elm Disease	*Ceratocystis ulmi*	4
Black Spot	*Gnomonia ulmea*	5
Miscellaneous Disorders		
Dieback	Stress factor(s)	1
Scorch	Heat, wind, and drought	1

ornamental problem (Table 3). The cause of dieback was related to a root and/or crown rot mostly commonly associated with poorly drained, wet sites in conjunc-

tion with *Phytophthora* spp. *Rhizoctonia* root rot was the most frequently recorded soilborne disease afflicting a variety of plants. Rust and twig blight were the main diseases found on juniper. Crown gall was frequently reported on spreading *Euonymus* during the early spring. Frequent summer rains enhanced a petal blasting caused by *Botrytis* sp. on a number of annual and perennial flowers, especially marigolds, peonies, and roses. Fungal and bacterial corm rots of gladiolus were common throughout the year. *Fusarium* stem canker and bacterial diseases were often found on greenhouse crysanthemum samples.

Disorders: Cold weather injury was common on many ornamental shrubs. Barberry showed extensive twig death and dieback due to cold injured stem tissue. Forsythia bloom was very sparse, occurring only on lower, snow covered twigs. Boxwood, *Azalea*, *Cotoneaster*, and *Pyracantha* were other shrubs that showed extensive cold damage. Scorch, decline, and dieback occurred on a number of ornamental species. The cause of these disorders was frequently related to poor site location, transplanting shock, and a variety of other stress factors (Table 3).

TABLE 3. *Ornamentals—Diseases and Disorders.*

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Ajuga (BUGLE WEED)		
Root Rot	*Rhizoctonia* sp.	1
Althaea (HOLLYHOCK)		
Rust	*Puccinia malvacearum*	1
Miscellaneous Disorder		
Leaf Curl	Environmental	1
Amaryllis (AMARYLLIS)		
Red Blotch	*Stagnospora curtisii*	1
Antirrhinum (SNAPDRAGON)		
Root Rot	*Rhizoctonia* sp.	1
Asarum (WILD GINGER))		
Root Rot	*Rhizoctonia* sp.	1
Miscellaneous Disorder		
Scorch	Environmental	1
Berberis (BARBERRY)		
Miscellaneous Disorder		
Dieback	Improper site/poor drainage	2
Boxus (BOXWOOD)		
Miscellaneous Disorder		
Winter Injury	Desiccation	1
Callistephus (CHINA ASTER)		
Aster Yellows	Virus	1
Wilt	*Fusarium oxysporum*	1
Miscellaneous Disorder		
Leaf Chlorosis	Environmental	1
Chlorophytum (SPIDER PLANT)		
Miscellaneous Disorder		
Root Rot	Improper cultural practices	1
Chrysanthemum (CHRYSANTHEMUM)		
Bacterial Blight	*Erwinia chrysanthemi*	1
Bacterial Leaf Spot	*Pseudomonas chicorrii*	2
Leaf Distortion	Chrysanthemum "B" virus	1
Miscellaneous Disorder		
Root Rot	Improper cultural practices	2

TABLE 3.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Cotoneaster (COTONEASTER)		
Twig Blight	*Cytospora* sp.	1
Leaf Spot	*Phyllosticta* sp.	1
Leaf Spot	*Botrytis* sp.	1
Crassula argentea (JADE PLANT)		
Miscellaneous Disorder		
Odema	Overwatered	1
Leaf Spot	Improper cultural care	1
Cydonia (QUINCE)		
Fire Blight	*Erwinia amylovora*	1
Cyperus (UMBRELLA PLANT)		
Root Rot	*Pythium* sp.	1
Dianthus (SWEET WILLIAM)		
Root Rot	*Rhizoctonia* sp.	1
Dieffenbachia (DIEFFENBACHIA)		
Miscellaneous Disorder		
Leaf Spot	Improper cultural care	2
Dracaena (DRACEANA)		
Miscellaneous Disorder		
Leaf Spot	Fluoride toxicity	1
Leaf Spot	Environmental	1
Euonymous (BURNING BUSH)		
Crown Gall	*Agrobacterium tumefaciens*	1
Leaf Spot	*Physllosticta* sp.	1
Miscellaneous Disorders		
Dieback	Stress factor(s)	3
Herbicide Injury	Spray drift	1
Adventitious Roots	Wet site	2
Euphorbia (GOPHER SPURGE)		
Root Rot	*Thaelaviopsis* sp.	1
Euphorbia pulcherrima (POINSETTIA)		
Crown/Stem Canker	Unidentified bacteria	1
Ficus (RUBBER PLANT)		
Miscellaneous Disorders		
Root Rot	Improper cultural conditions	1
Leaf Spot	Sunscald	2
Forsythia (GOLDEN BELLS)		
Miscellaneous Disorders		
Herbicide Damages	Spray drift	2
Dieback	Stress factor(s)/cold	1
Freesia (FREESIA)		
Corm Rot	*Fusarium* sp.	1
Gladiolus (GLADIOLUS)		
Corm Rot	*Fusarium oxysporum* f. *gladioli*	2
Corm Rot	*Curvularia trifolii* f. *gladioli*	1
Corm Rot	*Penicillium* spp.	3
Scab	*Pseudomonas marginata*	3
White Break Mosaic	Virus	
Miscellaneous Disorder		
Corm Rot	Storage breakdown	2
Hedera (ENGLISH IVY)		
Bacterial Leaf Spot	*Xanthomonas hederae*	2
Helianthus (SUNFLOWER)		
Miscellaneous Disorder		
Scorch	Environmental	1

TABLE 3.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Hemerocallis (DAYLILLY)		
Leaf Blight	*Kabatiella* sp.	1
Leaf Spot	*Heterosporium gracilis*	2
Hydrangea (HYDRANGEA)		
Flower Blight	*Botrytis*	1
Miscellaneous Disorder		
Decline	Stress factor(s)	2
Ilex (HOLLY)		
Berry Blight	*Botrytis* sp.	1
Miscellaneous Disorders		
Winter damage	Desiccation	2
Chlorosis	Iron deficiency	1
Scorch	Site Stress	1
Impatiens (IMPATIEN)		
Root Rot	*Rhizoctonia solani*	1
Ipomoea (MORNING GLORY)		
Miscellaneous Disorders		
Leaf Chlorosis	Nutrient imbalance	1
Scorch	Heat, wind, and drought	1
Iris (IRIS)		
Leaf Sopt	*Heterosporium iridis*	3
Crown Rot	*Botryotinia convoluta*	1
Basal Rot	*Fusarium* spp.	1
Juniperus (JUNIPER)		
Twig Blight	*Phomopsis juniperovora*	5
Cedar—Apple Rust	*Gymnosporangium juniperi-virginianae*	1
Twig Dieback	*Kabatina* sp.	1
Miscellaneous Disorders		
Dieback	Shading out/stress	4
Winter Damage	Desiccation	2
Ligustrum (PRIVET)		
Miscellaneous Disorders		
Dieback	Heat, wind, and drought	2
Winter Injury	Desiccation	2
Herbicide Damage	Spray drift	1
Narcissus (DAFFODIL)		
Leaf Blight	*Botrytis cinerea*	1
Orchid (ORCHID)		
Leaf Spot	Virus	1
Pachysandra (PACHYSANDRA)		
Leaf Blight	*Volutella pachysandrae*	2
Paeonia (PEONY)		
Botryis Blight	*Botrytis cinerea*	1
Parthenocissus (BOSTON IVY)		
Leaf Spot	*Guignardia bidwelli*	1
Pelargonium (GERANIUM)		
Blackleg	*Pythium* sp.	1
Bacterial Blight	*Xanthomonas pelargonii*	1
Leaf Spot	*Alternaria* sp.	1
Miscellaneous Disorder		
Oedema	Improper cultural conditions	1
Peperomia (PEPEROMIA)		
Miscellaneous Disorder		
Oedema	Improper cultural conditions	1

TABLE 3.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Petunia (PETUNIA)		
Root Rot	*Rhizoctonia* sp.	1
Philadelphus (MOCK ORANGE)		
Miscellaneous Disorders		
Canker	Stress factor(s)	1
Physocarpus (DWARF NINEBARK)		
Powdery Mildew	*Sphaerotheca macularis*	1
Polypodium (FERN)		
Miscellaneous Disorder		
Leaf Tip Burn	Low humidity	1
Polyscias fruticosa (MING ARALIA)		
Miscellaneous Disorder		
Chlorosis	Improper cultural conditions	1
Pothos (POTHOS)		
Miscellaneous Disorder		
Leaf Spot	Improper cultural conditions	1
Prunus (BUSH CHERRY)		
Miscellaneous Disorders		
Dieback	Stress factor(s)	1
Prunus (PLUM BUSH)		
Bacterial Leaf Spot	*Xanthomonas pruni*	1
Pyracantha (FIRETHORN)		
Scab	*Fusicladium pyracanthae*	3
Miscellaneous Disorder		
Winter Damage	Desiccation	2
Rhamnus (TALL HEDGE)		
Miscellaneous Disorder		
Leaf Scorch	Stress factor(s)	1
Rhododendron (AZALEA AND RHODODENDRON)		
Leaf Gall (Azalea)	*Exobasidium vaccinii*	1
Crown Rot (Rhododendron)	*Phytophthora* spp.	1
Miscellaneous Disorder		
Winter Scorch	Desiccation	3
Dieback	Transplant shock	3
Dieback	Mouse damage	1
Rosa (ROSE)		
Powdery Mildew	*Sphaerotheca* sp.	1
Blossom Blight	*Botrytis cinerea*	1
Miscellaneous Disorders		
Herbicide Injury	Spray drift	1
Leaf Scorch	Nutrient imbalance	1
Dieback	Poor graft union	2
Saintpaulia (AFRICAN VIOLET)		
Miscellaneous Disorder		
Root Rot	Soluble salt injury	1
Schefflera (AUSTRALIAN UMBRELLA TREE)		
Miscellaneous Disorder		
Oedema	Improper cultural conditions	2
Chemical Injury	Improper use	1
Root Rot	Overwatering	1
Senecio mikanioides (GERMAN IVY)		
Miscellaneous Disorder		
Dieback	Excess fertilizer	1

TABLE 3.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Syringa (LILAC)		
Leaf Spot	*Phyllosticta* sp.	1
Miscellaneous Disorder		
Small Leaves	Site stress	1
Tagetes (MARIGOLD)		
Leaf Spot	*Alternaria* sp.	2
Leaf Spot	*Septoria* sp.	1
Flower Blight	*Botrytis cinerea*	2
Miscellaneous Disorder		
Herbicide Injury	Spray drift	1
Taxus (YEW)		
Root Rot—Dieback	*Phytophthora*—poor drainage complex	14
Miscellaneous Disorder		
Herbicide Injury	Spray drift	2
Winter Damage	Desiccation	2
Dieback	Drought	2
Thuja (ARBORVITAE)		
Twig Blight	*Pestalotia funerea*	4
Miscellaneous Disorders		
Dieback	Transplant shock	2
Winter Injury	Desiccation	6
Inner Leaf Browning/Autumn	Natural phenomenon	2
Viburnum (VIBURNUM)		
Leaf Rust	*Coleosporium viburni*	1
Miscellaneous Disorder		
Dieback	Stress factor(s)	2
Vinca (PERIWINKLE)		
Stem/Leaf Blight	*Phoma exigua* var. *exigua*	1
Stem Canker	*Rhizoctonia* sp.	1
Yucca (YUCCA)		
Leaf Spot	*Coniothyrium concentricum*	1
Miscellaneous Disorder		
Leaf Spot	Improper cultural conditions	1

Tree Fruits

Diseases: Apple scab was not a problem in most orchards due to early dry spring weather (Table 4). As in previous years (1,2) fire blight was sporadic, with localized outbreaks ocurring in a number of orchards located in the southern half of the state. Blister spot of apple, a bacterial disease generally confined to the variety Mutsu, was observed for the first time on Stayman and Cortland varieties. The sudden increase in blister spot was attributed to wet weather which occurred shortly after petal fall. The most frequently recorded stone fruit disease was bacterial spot on peach, nectarine, apricot, and plum leaves (Table 4). Plum brown rot was reported from a number of backyard plantings.

Disorders: Severe winter cold periods killed peach, nectarine, and apricot fruit buds and resulted in a complete loss of those crops. Cold damage to trees was severe in many areas of the state. Many peach trees were killed while those that survived showed extensive death of individual stems and were late in leafing out. Leaf scorch on pear and apple caused by a combination of cold-injured tissue and dry, windy weather was common throughout the growing season. Russet-

TABLE 4. *Fruit Trees—Diseases and Disorders.*

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Citrus (ORANGE)		
Fruit Rot	Botrytis	1
Malus sylvestris (APPLE)		
Crown Rot	*Phytophthora cactorum*	1
Scab	*Venturia inaequalis*	1
Cedar-Apple Rust	*Gymnosporangium juniperi-virginianae*	2
Fireblight	*Erwinia amylovora*	4
Blister Spot	*Pseudomonas syringae*	2
Apple Blotch	*Phyllosticta* sp.	2
Bookers Disease	*Phoma* sp.	1
Pox	*Helminthosporium papulosum*	1
Miscellaneous Disorders		
Bud Death	Cold injury	1
Leaf Chlorosis	Nutrient imbalance	1
Measles	Manganese toxicity	1
Chemical Injury	Improper use	1
Adventitious Growth	Bur Knot	1
Twig Cankers	Hail	1
Scorch	Heat, wind, and drought	8
Fruit Russet	Environmental	1
Necrotic Leaf Blotch (Yellow Delicious)	Stress factor(s)	1
Bitter Pit	Physiological	4
Prunus americana (PLUM)		
Bacterial Leaf Spot	*Xanthomonas pruni*	3
Brown Rot	*Monilinia fructicola*	2
Black Knot	*Dibotryon morbosum*	3
Plum Pockets	*Taphrina deformans*	1
Miscellaneous Disorders		
Cold Injury	Low temperature	5
Canker	Animal damage	1
Chemical Injury	Improper use	1
Fruit Spot	Extreme heat	1
Prunus armeniaca (APRICOT)		
Bacterial Leaf Spot	*Xanthomonas pruni*	2
Miscellaneous Disorder		
Dieback	Stress factor(s)	3
Prunis avium (CHERRY)		
Miscellaneous Disorders		
Root Rot	Wet site location	4
Dieback	Stress factor(s)	7
Fruit Spot	Hail damage	4
Prunis persica (PEACH)		
Bacterial Spot	*Xanthomonas pruni*	3
Scab	*Cladosporium carpophilum*	1
Miscellaneous Disorders		
Root Rot	Wet side location	1
Herbicide Damage	Spray drift	1
Dieback	Cold injury	1
Chlorotic Leaves	Nutrient imbalance	1
Prunis persica var. *nectarina* (NECTARINE)		
Leaf Curl	*Taphrina deformans*	1
Bacterial Leaf Spot	*Xanthomonas pruni*	1
Pyrus communis (PEAR)		
Fire Blight	*Erwinia amylovora*	1
Scab	*Venturia pyrina*	1

TABLE 4.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Miscellaneous Disorders		
Decline	Cold injury	2
Scorch	Heat, wind, and drought	8
Chemical Injury	Improper use	4
Dieback	Boron deficiency	1

ting, a weather related skin blemish of golden delicious apples, was severe in most areas of the state. Also prominent on golden delicious was leaf yellowing and drop, a disorder thought to be caused by summer heat stress.

Small Fruits

Diseases: Strawberry root diseases were reported throughout Indiana in 1982 (Table 5). Black root rot, caused by an undetermined complex of soilborne pathogens, was frequently recorded during late summer. A contributing factor to the increased frequency of reports of black root rot was thought to be cold injury. Gray mold fruit rot of strawberry and brambles was severe in localized areas of the state due to cool, wet weather at harvest time and just prior to harvest. Cane blight, anthracnose and spur blight, fungal stem diseases, were widespread on brambles, especially red and black raspberries. The increase in bramble stem

TABLE 5. *Small Fruits—Diseases and Disorders*

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Fragaria grandiflora (STRAWBERRY)		
Black Root Rot	Fungal Complex (specific pathogens unknown)	4
Leaf Spot	*Mycosphaerella fragariae*	2
Gray Mold Rot	*Botrytis cinerea*	1
Slime Mold	*Physarum cinereum*	1
Red Steele	*Phytophthora fragariae*	2
Miscellaneous Disorders		
Scorch	Heat, wind, and drought	2
Root Rot	Cold injury/wet site	2
Wilt	Transplant shock	1
Rubus (RASPBERRY)		
Cane Blight	*Leptosphaeria coniothyrium*	2
Anthracnose	*Elsinoe veneta*	2
Rust	*Gymnoconia peckiana*	3
Spur Blight	*Didymella applanata*	1
Miscellaneous Disorders		
Poor Fruit Set/Decline	Cold injury	4
Vaccinium (BLUEBERRY)		
Twig Canker	*Phomopsis* sp.	2
Miscellaneous Disorder		
Dieback	Stress factor(s)	4
Vitis (GRAPE)		
Dead Arm Canker	*Phomopsis* sp.	1
Spot Anthracnose	*Elsinoe ampelina*	2
Miscellaneous Disorders		
Herbicide Injury	Spray drift	1
Poor Fruit Set	Cold	1
Poor Flowering	Stress factor(s)	1

diseases was due in part to cold injury which predisposed stem tissue to infection. Also noted was an increase in stem diseases of blueberries. Downy mildew of grape was severe on susceptible varieties in portions of southern Indiana during early summer. Powdery mildew of grape was observed during the fall.

Disorders: Strawberries and brambles were injured from the cold winter temperatures. Root and crown tissues of strawberries planted in exposed areas which had no snow cover or had not been mulched were extensively damaged. Numerous reports were received of strawberry plants which suddenly collapsed and died near harvest time. Cold injury symptoms on brambles appeared as a severe dieback of the plant and/or sudden collapse and withering of developing fruit. Such symptoms were most apparent on blackberries and non-hardy cultivars of red and black raspberries.

Turfgrasses

Diseases: *Helminthosporium* leaf spot and *Fusarium* blight were the most widespread and serious diseases of Kentucky bluegrass (Table 6). *Fusarium* blight and *Rhizoctonia* brown patch, coupled with excessive thatch and environmental stress, were responsible for killing large areas of turf in many lawns. Red thread (*Corticum fuciforme*) was diagnosed in several samples of bluegrass during the summer months and stripe smut (*Ustilago striiformis*), blackened lawns and caused some respiratory discomfort in late fall.

Disorders: Hot, dry weather resulted in poor root development and often caused plant death, especially where plants were further stressed by excessive thatch.

Vegetables

Vegetable Diseases/Disorders: As in previous years (1,2,3) specimens of cucurbit crops, soybean, tomato, and potato comprised the bulk of vegetable samples received (Table 7).

Diseases—Melons: Melons in Indiana were plagued by numerous problems in 1982. In southwestern Indiana, reports of *Fusarium* wilt were received shortly after

TABLE 6. *Turf—Diseases and Disorders.*

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Agrostis tenuis (BENTGRASS)		
Snowmold	*Typhula itoana*	2
Rhizoctonia (cool weather)	*Rhizoctonia* sp.	3
Lolium perenne (RYEGRASS)		
Miscellaneous Disorder		
Scorch	Improper cultural care	1
Poa pratensis (BLUEGRASS)		
Leaf Spot	*Helminthosporium* spp.	4
Fusarium Blight	*Fusarium roseum* complex	4
Stripe Smut	*Ustilago striiformis*	3
Rhizoctonia (warm weather)	*Rhizoctonia solani*	3
Red Thread	*Corticium fuciforme*	2
Fairy Ring	Basidiomycetes	1
Miscellaneous Disorders		
Chemical Injury	Improper use	1
Excessive Thatch	Improper cultural care	6
Scorch	Drought	4
Dieback	Nutrient deficiency	2

TABLE 7. *Vegetables—Diseases and Disorders.*

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Asparagus officinalis (ASPARAGUS)		
Needle Blight	*Cercospora asparagi*	1
Beta vulgaris (BEETS)		
Miscellaneous Disorder		
Wilt	Environmental stress	2
Barssica oleracea var. *acephala* (KALE)		
Miscellaneous Disorder		
Chlorosis	Nutrient imbalance	1
Brassica oleracea var. *botrytis* (CAULIFLOWER)		
Black Rot	*Xanthomonas campestris*	1
Head Blight	*Alternaria brassicae*	1
Brassica oleracea var. *capitata* (CABBAGE)		
Black Rot	*Xanthomonas campestris*	2
Miscellaneous Disorders		
Tip Burn	Physiological	2
Chemical Injury	Spray drift	1
Brasica ruvo (BROCCOLI)		
Head Blight	*Alternaria brassicae*	1
Wirestem	*Rhizoctonia solani*	1
Miscellaneous Disorder		
Leaf Spot/Oedema	Physiological	1
Capsicum frutescens (PEPPER)		
Wirestem	*Rhizoctonia solani*	1
Citrullus vulgaris (WATERMELON)		
Fusarium Wilt	*Fusarium oxysporum*	1
Leaf Spot	*Alternaria cucumerina*	1
Miscellaneous Disorders		
Scorch	Improper fertilizer application	1
Leaf Damage	Wind injury	2
Chlorosis	Nutrient deficiency	1
Cucumis melo (CANTALOUPE)		
Downy Mildew	*Pseudoperonospora cubensis*	1
Bacterial Wilt	*Erwinia tracheiphila*	4
Fusarium Wilt	*Fusarium oxysporum*	4
Miscellaneous Disorders		
Chlorosis	Nutrient imbalance	2
Leaf Damage	Wind injury	3
Chemical Injury	Spray drift	1
Scorch	Improper fertilizer application	1
Cucumis sativus (CUCUMBER)		
Bacterial Wilt	*Erwinia tracheiphila*	1
Angular Leaf Spot	*Pseudomonas syringae*	3
Miscellaneous Disorder		
Corky Rind	Environmental stress	1
Cucurbita moschata (PUMPKIN)		
Miscellaneous Disorder		
Chlorosis	Nutrient imbalance	1
Cucurbita pepo (ZUCCHINI)		
Mosaic	Cucumber mosaic virus	1
Miscellaneous Disorder		
Leaf Scorch	Heat, wind	2
Ipomoea batatas (SWEET POTATO)		
Scurf	*Monilochaetes infuscans*	1
Storage Rot	*Fusarium* sp.	1

TABLE 7.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Lactuca sativa var. *crispa* (LEAF LETTUCE)		
Miscellaneous Disorder		
Scorch	Environmental stress	1
Lycopersicon esculentum (TOMATO)		
Septoria Leaf Spot	*Septoria Lycopersici*	14
Early Blight	*Alternaria solani*	4
Verticillium Wilt	*Verticillium albo-atrum*	1
Black Dot Root Rot (Hydroponics)	*Colletotrichum coccodes*	1
Southern Wilt	*Sclerotium rolfsii*	1
Bacterial Spot	*Xanthomonas vesicatoria*	1
Bacterial Speck	*Pseudomonas syringae*	3
Anthracnose	*Colletotrichum coccodes*	1
Double Streak Virus	Tobacco Mosaic Virus + Potato Virus X	1
Gray Mold (Hydroponics)	*Botrytis* sp.	2
Mosaic	Cucumber Mosaic Virus	1
Mosaic	Tobacco Mosaic Virus	1
Miscellaneous Disorders		
Chemical Injury	Spray Drift	7
Chlorosis	Nutrient imbalance	3
Purpling	Phosphorous deficiency	1
Blossom End Rot	Physiological	1
Walnut Wilt	Juglone toxin	2
Poor Fruit Set	Improper pollination	1
Stem Canker	Hail	1
Catface	Environmental stress	3
Leaf Curl/Necrosis (Hydroponics)	Nutrient imbalance	4
Corky Root	High soluble salt level	1
Fruit Injury	Sunscald	2
Leaf Roll	Physiological	4
Leaf Spot (Hydroponic)	Improper management	2
Fruit Rot	Excess water	1
Phaseolus lunatus (LIMA BEAN)		
Miscellaneous Disorder		
Leaf Scorch	Heat, wind	1
Phaseolus vulgaris (SNAP BEAN)		
Root Rot	*Rhizoctonia solani*	1
Root Rot	*Fusarium* sp.	2
Anthracnose	*Colletotrichum lindemuthianum*	1
Angular Leaf Spot	*Isariopsis griseola*	2
Stem Rot	*Sclerotinia sclerotiorum*	3
Rust	*Uromyces phaseoli*	1
Mosaic	Cucumber Mosaic Virus	1
Miscellaneous Disorders		
Chemical Injury	Spray Drift	2
Leaf/Root Burn	Fertilizer burn	3
Leaf Scorch	Wind injury, sunscald	11
Pisum sativum (PEA)		
Miscellaneous Disorder		
Chemical Injury	Spray drift	1
Raphanus satijus (RADISH)		
Scab	*Streptomyces scabies*	1
Rheum spp. (RHUBARB)		
Anthracnose Leaf Spot	*Colletotrichum erumpens*	1
Miscellaneous Disorder		
Scorch	Site stress	1

TABLE 7.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Solanum tuberosum (POTATO)		
Tuber Rot	*Fusarium* sp.	1
Early Blight	*Alternaria solani*	2
Common Scab	*Streptomyces scabies*	1
Verticillium Wilt	*Verticillium albo-atrum*	1
Miscellaneous Disorders		
Leaf Curl	Herbicide drift	1
Seed Piece Decay	Normal	1

muskmelons and watermelons were transplanted. Yield reduction in several of the more seriously affected fields was estimated to be more than 50%. Incidence of bacterial wilt of muskmelon was greater in 1982 than in previous years. The increase was attributed to an unusually large and early increase in striped cucumber beetle populations. Powdery mildew was observed throughout the state, but its effect on yield was minimal because disease became established relatively late in the season. Downy mildew, anthracnose, and gummy stem blight were also reported in 1982. A few severe cases of these diseases were observed, but the statewide incidence was generally very low. The most troublesome foliage disease was *Alternaria* leaf blight. This disease occurred wherever less vigorous plants were grown. Stress that predisposed muskmelons to *Alternaria* could be attributed to several factors, including the presence of other diseases, extremes of high or low water availability, and nitrogen deficiency caused by leaching or excessive fruit load.

Disorders: A foliage blight that appeared in many fields where sandy ridges existed has been linked to a deficiency of available magnesium in the soil. Symptoms are somewhat similar to those of downy mildew, but more closely resemble descriptions of magnesium deficiency.

Diseases—Tomatoes: Tomatoes in commercial fields and home gardens were infected by early blight throughout most of the season. The occurrence of stem lesions increased the severity of disease development in 1982. Where stem lesions were established early, very thin stands resulted. Stem lesions that developed later in the season weakened plants to the extent that foliage was more prone to infection by *Alternaria*. This usually resulted in premature ripening and dropping of fruit and increased sunscald. Higher incidence of fruit lesions and anthracnose usually accompanied severe early blight defoliation. *Septoria* leaf spot was commonly diagnosed on plants suffering from early blight. Bacterial spot and speck presented problems in a few commercial fields early in the season. Reports of bacterial canker were rare, and, because symptoms appeared late in the season, yield loss to canker was negligible.

Disorders: As in previous years (1,2), a high number of tomato samples were diagnosed as having chemical injury (spray drift). Environmental factors, such as sunscald and water fluctuations caused foliage and fruit disorders. Nutrient deficiencies and physiological disorders were also diagnosed.

Diseases—Other Vegetable Crops: Black rot, caused by *Xanthomonas campestris* was rampant among crucifers of all types (cabbage, broccoli, cauliflower, and Brussels sprouts). Only varieties most resistant to black rot escaped damage. Many garden plantings and several commercial fields were completely destroyed.

Phytophthora blight of pepper and *Verticillium* wilt of okra and eggplant were common (some cases were very severe) in northwestern Indiana.

Although onion purple blotch, caused by *Alternaria pori* was frequently observed in muck areas of northern Indiana, damage was generally slight.

Agronomic Crops

Agronomic Diseases/Disorders: Damaging weather conditions were directly or indirectly related to an increased severity of several diseases and disorders of agronomic crops (Table 8).

TABLE 8. *Agronomic Crops—Diseases and Disorders*

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Triticum (WHEAT)		
Take-All	*Ophiobolus graminis*	6
Fusarium Root Rot	*Fusarium* spp.	4
Wheat Spindle Streak	Wheat Spindle Streak Virus	2
Barley Yellow Dwarf	Barley Yellow Dwarf Virus	3
Spring Blight	*Ceratobasidium* spp.	6
Leaf Rust	*Puccinia recondita* f. sp. *tritici*	
Stem Rust	*Puccinia graminis* f. sp. *tritici*	
Septoria Leaf Blotch	*Septoria tritici*	2
Septoria Glume Blotch	*Septoria nodorum*	2
Helminthosporium Leaf Spot	*Helminthosporium sativum*	1
Tan Spot	*Pyrenophora trichostoma*	2
Scab	*Gibberella zeae*	6
Miscellaneous Disorders		
Improper Root Development	Broadcast planting onto crop residue	6
Chlorosis	Nutrient imbalance	4
Chlorosis	Drought	1
Avena (OAT)		
Barley Yellow Dwarf	Barley Yellow Dwarf Virus	7
Miscellaneous Disorders		
Chlorosis	Chemical injury	1
Improper Root Development	Broadcast planting onto crop residue	1
Leafspot	Physiogenic	1
Mold	Improper drying	1
Elymus (RYE)		
Ergot	*Claviceps-purpurea*	1
Zea (DENT CORN)		
Gibberella Seedling Blight	*Gibberella zeae*	1
Anthracnose Leaf Blight	*Colletotrichum graminicola*	14
Northern Corn Leaf Spot	*Bipolaris carbonum* (Race II)	10
Southern Leaf Blight	*Bipolaris maydis* (Race 0)	12
Common Leaf Rust	*Puccinia sorghi*	10
Stewart's Blight	*Erwinia stewartii*	2
Common Smut	*Ustilago maydis*	1
Fusarium Kernel Rot	*Fusarium moniliforme*	2
Gibberella Kernel Rot (grown 1981)	*Gibberella zeae*	40
Gibberella Kernel Rot (1982)	*Gibberella zeae*	1
Pythium Seedling Blight	*Pythium aphanidermatum*	2
Kernel Rot	*Aspergillus glaucus*	2
Kernel Rot	*Aspergillus flavus*	1
Kernel Rot	*Cephalosporium* sp.	1
Kernel Rot	*Diplodia zeae*	2
Fusarium Stalk Rot	*Fusarium moniliforme*	2
Gib Stalk Rot	*Gibberella zeae*	4
Helminthosporium Stalk Rot	*Bipolaris carbonum*	1
Anthradnose Stalk Rot	*Colletotrichum graminicola*	6
Fusarium Stalk Rot	*Fusarium* spp.	2

TABLE 8.—Continued

Host Plant Diseases and/or Disorders	Causal Agent	Number of Samples
Miscellaneous Disorders		
Fungal Growth (Rotted Stalk)	*Typhula* sp.	2
Leaf Purpling	Environmental stress	4
Leaf Spot	Paraquat spray drift	1
Root Restriction/Stunting	Soil compaction	4
Reduced Root Growth	Chemical Injury	4
Leaf Spot	Physiogenic	1
Leaf Scorch	Chemical Injury	3
Wilt	Drought	1
Chlorosis	Environmental stress	3
Rolled in Shoot	Herbicide/mechanical damage	3
Gylcine (SOYBEAN)		
Rhizoctonia Root Rot	*Rhizoctonia solani*	15
Phytophthora Root Rot	*Phytophthora megasperma* var. *sojae*	6
Pythium Root Rot	*Pythium aphanidermatum*	1
Bacterial Blight	*Pseudomonas gylcinea*	2
Pod and Stem Blight	*Diaporthe phaseolorum* var. *sojae*	2
Brown Stem Rot	*Cephalosporium gregatum*	2
Anthracnose	*Collectotrichum graminicola*	1
Charcoal Rot	*Macrophomina phaseolina*	4
Brown Spot	*Septoria glycines*	12
Soybean Cyst Nematode	*Heterodera gylcines*	4
Stem Rot	*Sclerotinia sclerotiorum*	4
Downy Mildew	*Peronospora manshurica*	4
Miscellaneous Disorders		
Chemical Injury	Various causes	20
Chlorosis	Nutrient deficiency	1
Root Restriction	Soil compaction	2
Leaf Discoloration	Sunscald	2
Stunted Growth	Environmental factors	3
Thick Hypocotyl	Planted too deep	1
Trifolium (CLOVER)		
Mosaic Virus	Clover Mosaic Virus	2
Root Rot	*Fusarium* spp.	1
Medicago sativa (ALFALFA)		
Root Rot	*Phytophthora megasperma*	5
Downy Mildew	*Peronospora trifoliorum*	1
Root Knot Nematode	*Meloidogyne*	1
Spring Blackstem	*Phoma medicaginis*	1
Summer Blackstem	*Cercospora medicaginis*	1
Leptosphaerulina Leaf Spot	*Leptosphaerulina briosiana*	3
Crown Root Rot	Fungal complex	6
Common Leaf Spot	*Pseudopezizza medicaginis*	5
Stemphylium Leaf Spot	*Stemphylium botryosum*	3
Rust	*Uromyces striatus*	1
Miscellaneous Disorders		
Slow Growth	Nutrient imbalance	2
Leaf Discoloration	Environmental factors	2
White spots on leaves/curling	Frost	1
Leaf Purpling	Boron deficiency	2
Stunting	Compaction	2
Nicotiana (TOBACCO)		
Miscellaneous Disorder		
Leaf Spot	Nutrient imbalance	1

Diseases—Wheat: Significant yield losses occurred from infectious diseases in 1982, but yield losses from disease were not as great as in 1981 even though state

average yields were much lower. Powdery mildew and *Septoria* leaf blotch of wheat were prevalent diseases throughout the state, but rarely did they advance to the point of killing the flag leaf. Leaf rust, which appeared earlier than usual, was very widespread. However, severe levels of infection did not develop until late in the season and therefore, yield losses were relatively slight. *Septoria* glume blotch and head scab were widespread and significantly contributed to reduced yields and reduced seed quality. *Rhizoctonia* spring blight was found in many areas of the state. The disease may be easily confused with winter killing, and therefore it may have been more widespread than indicated. Barley Yellow Dwarf Virus was widespread in the state. Take-all was much more evident in 1982 than in 1981 and was more prevalent in the northern part of the state.

Disorders: Winter killing of wheat was a significant problem in Indiana during 1981-82. The damage was especially severe in the northeastern quarter of the state, and many fields in this area were plowed and planted to another crop.

Disease—Oats: Barley Yellow Dwarf Virus was especially damaging to oats in the northern half of the state.

Diseases - Soybeans: Soybean seedling blights caused by *Rhizoctonia*, *Pythium*, and *Phytophthora* were widespread. *Rhizoctonia* seedling blight and later root rot were probably more severe this year than previously reported. *Septoria* brown spot caused defoliation of soybeans in some fields early in the growing season. In most areas of the state, dry conditions during August reduced the potential yield losses from this disease. Downy mildew was widespread in 1982 and some seed infection was observed. Charcoal root rot was extensive and, in many cases, severe in the southwestern quarter of the state. Estimated yield losses of 10 to 15 bushels per acre from this disease were common in Gibson County. *Sclerotinia* stem rot was observed in scattered individual fields in the central and northeastern portion of the state for the second successive year (2). Brown stem rot was commonly found in the northern half of the state. *Sclerotinia* and brown stem rot significantly reduced yields in those fields where the disease occurred. Injury from the soybean cyst nematode continued to increase in frequency and severity. Occurrence of the nematode was documented in Crawford County for the first time. Charcoal root rot, *Sclerotinia* stem rot, brown stem rot, and soybean cyst nematode pose a threat to soybean production in Indiana in the future years.

Diseases—Corn: Leaf blights of corn were common throughout the state during 1982, but yield losses from these diseases were minimal. Early season anthracnose leaf blight was more severe in eastern Indiana than in any previous year. In many cases the disease killed the first four to six leaves of plants. By the time the plants reached the six to eight leaf stage, disease progression decreased markedly. Southern corn leaf blight, *Kabatiella* eye spot, northern corn leaf blight (races 1 and 2), northern corn leaf spot, and common rust were diseases commonly found in Indiana corn fields in 1982. However, yeld losses from these diseases were minimal. The top kill phase of anthracnose was observed throughout the state. Stalk rots became severe in many fields as corn reached maturity. *Gibberella* and *Fusarium* stalk rots were common throughout the state. Charcoal stalk rot was severe in some southwestern Indiana fields, and *Diplodia* stalk and ear rots were severe in some fields in Orange and Washington counties. *Diplodia* stalk and ear rots appeared to be severe only in those fields where corn followed corn and where reduced tillage operations were used. Ear rots were generally very light in 1982, although in addition to the individual cases of *Diplodia* ear rot, some minor *Gibberella* and *Fusarium* ear rots were observed.

Disorders: Vivipary (germination of corn kernels *in situ*) was observed in three

widely separated fields. The cause of this malady is unknown. Environmental stress caused numerous cases of leaf purpling, spotting, and chlorosis (Table 8). At one point during early summer, corn plants in several different fields exhibited rolling of the leaves. Possible causal factors were listed as environmental, chemical, and herbicidal stress. However, no precise diagnosis was possible.

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L-Methionine Sulfoximine as a New Electron Acceptor in Photosystem I of Spinach Chloroplasts

THOMAS W. SENG, RITA BARR and FREDERICK L. CRANE
Department of Biological Sciences
Purdue University, West Lafayette, Indiana 47907

Abbreviations: DCMU-3-(3,4-dichlorophenyl)-1,1-dimethylurea; DBMIB-2, 5-dibromo-3-methyl-6-isopropyl-p-benzoquinone or dibromothymoquinone; MSO-L-methionine sulfoximine; MV-methylviologen; PSI-photosystem I.

Introduction

Methylviologen, the best known electron acceptor in Photosystem I of spinach chloroplasts, or other viologen-type compounds presumably accept electrons from the reaction center complex or from a bound iron-sulfur protein associated with this complex (1). The purpose of this study was to look for other artificial electron acceptors in the methylviologen region and to find unique inhibitors, which would differentiate between these pathways. L-Methionine sulfoximine proved to be such an electron acceptor, capable of differentiating a site after the methylviologen site. Such a conclusion was reached on the basis of using 1-(2-thiazolylazo)-naphthol as a specific inhibitor for the new pathway.

Materials and Methods

Chloroplasts were isolated from market spinach in 0.4 M sucrose and 0.05 M NaCl as previously described (2). Chlorophyll was determined according to Arnon (3). Oxygen uptake was measured with a Clark-type oxygen electrode according to Barr and Crane (2). Reaction mixtures for assays are given in figure legends. Reaction rates were recorded with a Sargent-Welch SRG recorder.

Results and Discussion

L-methiomine sulfoximine gives oxygen uptake like methylviologen when used as an electron acceptor in PS I. As Table I shows, optimum reaction rates were obtained with a concentration between 50 and 100 μM at pH 7. The $H_2O \rightarrow$

Assay Conditions for the Photosystem I Reaction
H_2O → Methionine Sulfoximine (+ Na Azide)

Methionine Sulfoximine (μM)	Reaction Rate (μequiv. O_2/mg chl•hr)
I Optimum Concentration	
25	192
50	226
75	395
100	338
75 (minus Na azide)	68
75 (minus NH_4Cl)	90
II Optimum pH	
pH 6	297
pH 7	395
pH 8	221

Inhibition of the H_2O → Methionine Sulfoximine Reaction by Standard Inhibitors

Inhibitor	Conc.	Electron Transport H_2O → MV (+ azide), pH 7		H_2O → MSO (+ azide), pH 7	
		Rate[a]	Inhibition	Rate[a]	Inhibition
	(μM)		(%)		(%)
None	—	538	0	310	0
DCMU	5.0	0	100	0	100
DBMIB	2.5	56	90	56	82
Bathophenanthroline	100	60	89	42	86
Polylysine (M.W. 30,000)	50 μg/ml	96	82	62	80

[a] μequiv. O_2mg chl•hr

methylviologen (+ azide) and the H_2O→ methionine sulfoximine (+ azide) reactions were inhibited to a similar extent by the standard inhibitors for sites between the 2 photosystems: DCMU for the B protein site (4), dibromothymoquinone for the plastoquinone pool (4), bathophenanthroline for the Rieske iron-sulfur protein located between the plastoquinone pool and cytochrome f (5), and polylysine for plastocyanin (6). These inhibitions range from 92-100% for both reactions (Table II). Ions, such as $Ca^{2}+$, $Mg^{2}+$, $Sr^{2}+$, or $Ba^{2}+$ stimulate both reactions (Fig. 1 and 2), although to a lesser degree on the H_2O→ methionine sulfoximine pathway.

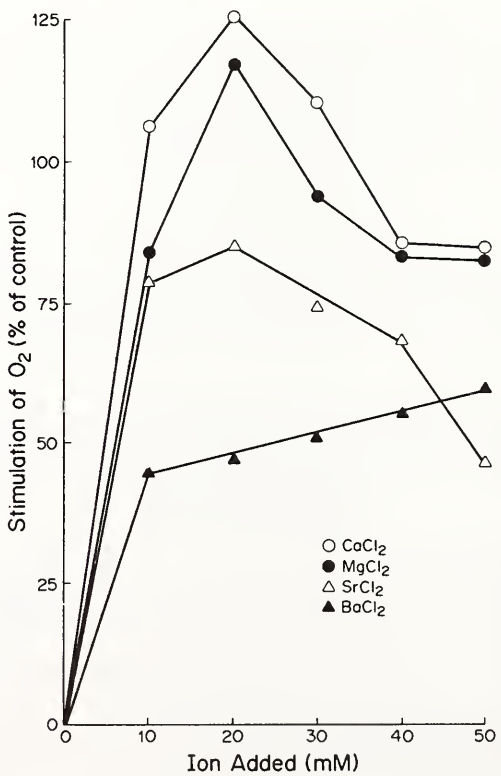

FIGURE 1. *Stimulation of Electron Transport in Spinach Chloroplasts by Ions. The reaction tested was H_2O→ methylviologen (+azide), pH 7. The reaction mixture contained chloroplasts (0.05 mg. chlorphyll), 25 mM Tris-Mes, pH 7, 5mM NH_4Cl, 0.5 mM Na azide, and 0.5 mM MV. The control rate was 530 uequivalents O_2/mg chl•hr.*

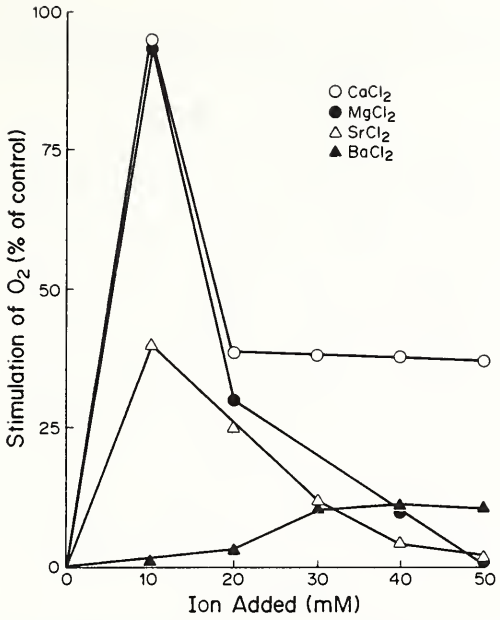

FIGURE 2. *Stimulation of Electron Transport in Spinach Chloroplasts by Ions. The reaction tested was $H_2O \rightarrow$ methionine sulfoximine (+azide), pH 7. The reaction mixture contained chloroplasts (0.05 mg chlorophyll), 25 mM Tris-Mes, pH 7, 5mM NH_4Cl, 0.5 mM Na azide, and 75 μM MSO. The control rate was 281 μquiv. O_2/mg chl•hr.*

The data presented so far allow no differentiation between the two pathways. Only the use of such an iron chelator as 1-(2-thiazolylazo)-naphthol (Fig.3) or incubation with sodium selenite (Fig. 4) uncovers differences. As Fig. 3 shows, the iron chelator inhibits the $H_2O \rightarrow$ methionine sulfoximine pathway more than 80%,

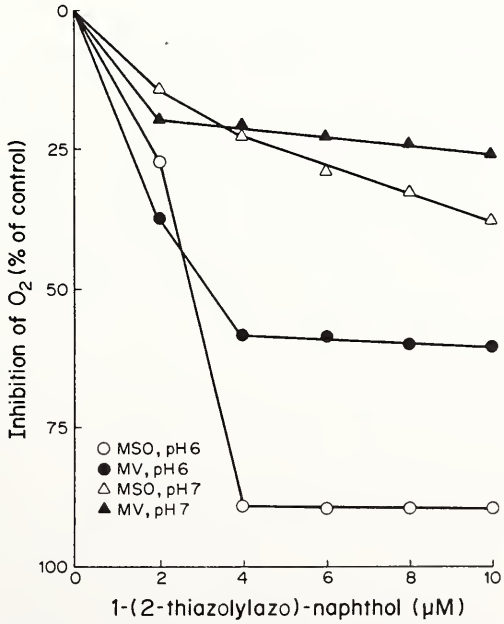

FIGURE 3. *The Inhibition of Electron Transport in Spinach Chlorophasts by 1-(2-Thiazolylazo)-Naphthol. The control rate for $H_2O \rightarrow MV$ (+azide), pH 7 was 575 μequiv. O_2/mg chl•hr, for $H_2O \rightarrow MSO$ (+azide), pH 7, it was 293. The reaction mixtures were as in Figures 1 and 2.*

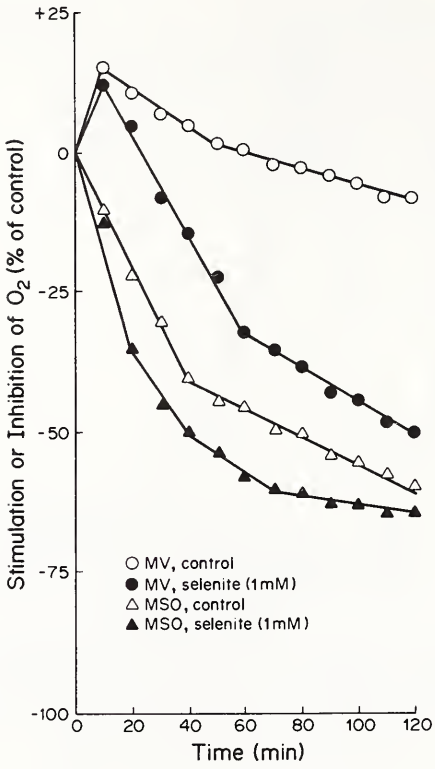

FIGURE 4. *The Inhibition of Electron Transport in Spinach Chloroplasts by Sodium Selenite. The control rate for $H_2O \rightarrow MV$ (+azide), pH 7 was 496 μequiv. O_2mg chl•hr, for $H_2O \rightarrow MSO$ (+azide), pH 7 it was 338. The reaction mixture were as Figures 1 and 2.*

but the $H_2 \rightarrow$ methylviologen pathway over 50%. This indicates that the new electron acceptor methionine sulfoximine is able to reach a site past where methylviologen accepts electrons in PS I. This new site may be a bound iron-sulfur protein, several of which have been detected by studies of PS I reaction center particles by flash-induced absorption changes (7,8) or by EPR spectroscopy (9). With the idea in mind that selenium ions can replace sulfur in iron-sulfur protein cores (10), a 120-min. incubation of chloroplasts with sodium selenite was undertaken. As Fig. 4 shows, the $H_2O \rightarrow$ methylviologen reaction was inhibited about 50% in two hours, but the $H_2O \rightarrow$ methionine sulfoximine pathway was inhibited only about 5% compared to its sucrose-NaCl control. However, since the methionine sulfoximine pathway and its control lost about 60% activity, it is possible that the acceptor site reached by methionine sulfoximine is an iron-sulfur site exposed to the outside of the membrane, which has lost its labile sulfur during incubation, while exposed to atmospheric oxygen. A total of 3-4 bound iron-sulfur centers are known to exist in PS I (11). It is also known that the sulfur in iron-sulfur clusters is labile, particularly when exposed to atmospheric oxygen (12). The fact that selenite was able to inhibit the $H_2 \rightarrow$ methylviologen pathway 50%, implies that its iron-sulfur was originally less exposed to the outside than the presumed iron-sulfur on the $H_2O \rightarrow$ methionine sulfoximine pathway.

In conclusion, it can be seen that L-methionine sulfoximine can act as an electron acceptor in PS I and that it may accept electrons at a different point that methylviologen. This site is more sensitive to loss of activity in contrast to the selenium-inhibited iron-sulfur site found on the methylviologen pathway.

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CELL BIOLOGY

Chairman: Douglas W. Duff
Department of Biology, Indiana University at South Bend,
South Bend, Indiana 46634

Chairman-Elect: Zafar Iqbal
School of Medicine, Indiana University Indianapolis, Indiana 46240

ABSTRACTS

Role of Calmodulin in Nerve Function. Z. Iqbal, L. McKitrick, and S. Ochs, Department of Physiology, School of Medicine, Indiana University, Indianapolis, Indiana 46223.—— Calcium plays an important role in a number of physiological functions and recently it has been demonstrated that the effects of calcium are mediated through a calcium-binding protein calmodulin. Calmodulin, a heat-stable acidic protein of about 17-18K Dalton has been found present in a variety of tissues isolated from various sources. We have recently isolated and characterized the protein from mammalian nerve fibers. This communication describes the possible role of this protein in an important nerve cell function, i.e., the axoplasmic transport. Axoplasmic transport is required for the supply of needed materials in the nerve cell processes for the maintenance and normal physiological activities. The study of axoplasmic transport in our laboratory is conducted with the help of a radioactive tracer technique. The radioactive labeled aminoacids are injected into the L-7 dorsal root ganglion of cat and the sciatic nerve is removed from the cat after an insitu transport of 2 hr. The peroneal branch is desheathed to expose it to the invitro medium while the tibial branch is left intact to act as control. After a determined time interval the nerves are assessed for the radioactivity. Employing these procedures we have found that depletion of calcium from the medium or the incorporation of calmodulin-inactivating agents like trifluoperazine blocks the transport of the labeled proteins in the peroneal branch of the nerve which was exposed to these agents thus suggesting that the transport is dependent on calcium and that the effects of calcium are likely to be mediated through calmodulin.

The Effect of Hyperthermia on the Murine Mammary Tumors: A Light and Electron Microscopic Study. Mohinder S. Jarial and Duncan T. Kennedy, Department of Physiology and Health Science and Muncie Center for Medical Education, Ball State University, Muncie, Indiana 47306.—— Renewed interest in treating malignant tumors with hyperthermia led to this investigation of structural changes in murine mammary tumors following hyperthermia. Female, Strong A mice, which have a high incidence of mammary tumors, were anesthetized with Nembutal (40 mg/Kg, i.p.). Tumors were removed at 5, 20 minutes, 24, 48, 72 hours and 7 days after localized heating for one hour via hot water (47.5°C) circulated through a plastic bag.

A significant EM observation in untreated tumor cells was presence of large numbers of virus particles in virtually all Strong A mouse tumor cells. These particles, larger in acinar lumina and spaces than in cytoplasm, had membrane bound dense cores. Treated cells showed nuclear membrane thickening, cytoplasmic vacuolization and mitochondrial disruption at 5 minutes posthyperthermia. At 24 hours tumor cells were separating, their nuclei showed fragmented, clumped

chromatin with blebs and breaks in the membrane. Acid phosphatase-positive lysosomes containing dense bodies and virus particles were most prominent at 24 hours. After 24 hours tumor cells showed severe degenerative changes with loss of cell boundaries and cytoplasmic organelles.

This study confirms the effectiveness of hyperthermia in destroying malignant tumor cells *in vivo* and suggests that increased lysosomal activity with damage to the nucleus and cytoplasmic organelles are critical in the process. Supported by a grant from Delaware County Cancer Society.

Superoxide Dismutase Activity in Crown Gall Tumors. JULIE A. OLSEN and MARK HATFIELD, Wabash College, Crawfordsville, Indiana 47933.—— Numerous studies of mammalian tumor cells have shown these cells to have reduced levels of Mn superoxide dismutase (MnSOD). This enzyme deficiency in mammalian tumor cells is thought to lead to increased levels of superoxide which causes further cell damage. It is of interest to determine whether this is a common feature of both mammalian and plant tumor cells. Of particular interest is the crown gall tumor which is currently being explored as a screening system for antineoplastic agents.

Three different strains of agrobacterium tumefaciens were used to generate crown gall tumors in etiolated decapitated pea stems. The soluble superoxide dismutase activity in individual pea stem tumors was compared with untransformed etiolated decapitated pea stems. The MnSOD activity did not diminish due to tumor cell transformation. Variations in the MnSOD activity appear to be correlated with the length of the side shoots produced on the stem. The CuZn SOD activity varied widely; however, the CuZn SOD activity in the tumor cells did not fall below that in the normal tissue.

Effects of Dietary Stearic Acid on Cellular Characteristics of Mouse Mammary Tissue and Spontaneous Mammary Adenocarcinomas. M.L. RICHESON, Assistant Professor of Biology, I.U. at Purdue, at Fort Wayne, Fort Wayne, Indiana 46805 and A.S. BENNETT, Department of Biology, Ball State University, Muncie, Indiana 47306.—— High fat diets have been implicated in the production of mammary adenocarcinomas, but mechanisms involved are not clear. Strong Strain A female mice maintained on a 15% fat diet (13 + % stearic acid) develop spontaneous mammary adenocarcinomas at a later age than those fed Purina Rodent Chow (4% fat). Dietary fatty acids have a significant effect on the fatty acid distribution of tumorous tissue and plasma membrane composition of mammary tissue.

This study was conducted to compare effect of dietary stearic acid on morphological characteristics of normal and tumorous mammary tissue. Particular emphasis was placed on identifying the distribution and localization of lipid in cell structure of tissues removed from animals on high and low fat diets.

Retinol Enhances Aggregation and Fusion of Egg Lecithin Liposomes. WILLIAM STILLWELL and STEVEN NAHMIAS, Department of Biology, Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana 46223.—— Previously we have demonstrated that retinoids, vitamin A and related compounds, can affect several physical parameters of lipid bilayers (Stillwell, Ricketts, Hudson, Nahmias, 1982, Biochim. Biophys. Acta 688: 653-659). Retinol and retinoic acid both increase membrane permeability to cations, anions, neutral solutes and water, increase electrical conductance of planar bimolecular lipid membranes and lower the phase transition temperature of synthetic phosphatidylcholine membranes. Here we report a retinol-dependent enhancement of aggregation and fusion of egg lecithin liposomes. Aggregation is followed spectrophotometrically by changes in absorbance or light scattering while fusion is demonstrated by a sensitive fluorimetric

technique employing mixed populations of terbium-containing liposomes and dipicolinic acid-containing liposomes. Rates of aggregation and fusion are increased by retinol when incorporated into the lipid bilayers at levels from 1 to 10 membrane mole percent. The significance of these results in elucidating the systemic, non-visual role for vitamin A is discussed.

Preparation of Liposomes and Their Use in Studies with Paramecium

THOMAS A. COLE, CHRISTOPHER C. SCHROEDER, and WILLIS H. JOHNSON
Department of Biology, Wabash College, Crawfordsville, Indiana 47933

Liposomes are synthetic lipid vesicles, first described in 1965 by Bangham, et al. (1, 6). The major interests in liposomes are as models of biological membranes and as exogenous delivery systems for targeting of encapsulated agents, such as deoxyribonucleic acid (DNA), drugs or enzymes to specific tissues. Methods for preparation of liposomes are varied (12): sonication of dried lipid films in presence of aqueous solution, reverse phase evaporation, extrusion through a small orifice (French press), ethanol injection into aqueous media, detergent dialysis (7, 9), gel filtration (4), and transient pH changes (2).

Here we report preparation of liposomes by simple autoclaving, the use of primulin for visualization of liposomes in a fluorescent microscope, and the use of CHAPS detergent for liposome preparation in the detergent dialysis method.

Materials and Methods

Chloroform and diethyl ether were reagent grade and were used without further purification. β-Octyl glucoside and tocopherol were from Sigma Chemical Company (St. Louis, MO). Primulin (Direct Yellow 59) was from Aldrich Chemical Company (Milwaukee, WI). 11-(9-anthroyloxy)-undecanoic acid, 2-(9-anthroyloxy)-palmitic acid, cholesteryl anthracene-9-carboxylate, and dansyldipalmitoyl-L-α-phosphatidyl ethanolamine (dansyl-DPPE) were from Molecular Probes, Inc. (Junction City, OR). CHAPS detergent (3-((3-Cholamidopropyl)-dimethylammonio)-1-propanesulfonate) was from PolySciences, Inc. (Warrenton, PA) or Calbiochem-Behring, Inc. (La-Jolla, CA). Dipalmitoylphosphocholine, a synthetic phospholipid was obtained from Molecular Probes, Inc. or Sigma. Dialysis tubing was Spectrapor, 1/4" diameter, molecular weight cutoff = 12,000 from Arthur H. Thomas Company (Philadelphis, PA).

Results

1. Preparation of Liposomes by Autoclaving

Methods for reverse phase evaporation of volatile organic solvents for liposome preparation involve dissolving the phospholipid in chloroform with a neutral lipid such as tocopherol and mixing this solution with the same amounts of lipids dissolved in diethyl ether before evaporation under a stream of nitrogen gas (5, 10). We have found that the chloroform and ether may be removed quickly by autocalving the mixture at 15 psi for 12 minutes. Many large liposomes are formed. If none of the components for liposome preparation are heat labile, autoclaving compares favorably to nitrogen-induced evaporation in size and numbers of liposomes produced. For example, 2.4 mg (3.33 μmol) DPPC and 1 drop of tocopherol are dissolved in 1 ml of chloroform and 1 ml of diethylether separately. One ml 50 mM KC1 is added to the chloroform solution and 5 ml 50 mM KC1 is added to the ether solution. Both solutions are shaken vigorously, mixed together and shaken again before autoclaving. The resulting liposomes are mainly in the 0.5 to 20 μ diameter range. The cylindrical-type autoclave used is from American Sterilizer Co. (Erie, PA) with an inner chamber volume of 6.95 1 (2.45 ft^3, diameter = 15 in, depth = 24 in) and electrical heat. We experienced no difficulties in autoclaving 1 ml of ether and 1 ml of chloroform in an autoclave

of this size and think that the procedure is not hazardous. However, potential hazards should be recognized.

We have prepared fluorescent liposomes by adding a fluorescent lipid to the DPPC-tocopherol in chloroform and ether solutions. 11-(9-anthroyloxy)-undecanoic acid, 2-(9-anthroyloxy)-palmitic acid, and cholesteryl anthracene-9-carboxylate all are incorporated into the synthetic membrane uniformly.

We also have prepared fluorescent liposomes from a fluorescent phopholipid, dansyl-DPPE, without DPPC. This shows that fluorescent liposomes can be prepared from fluorescent phospholipids as well as by intercalation of fluorescent fatty acid and cholesterol derivatives into the DPPC bilayer.

2. Use of Primulin to Visualize Liposomes

Primulin has been used as a spray reagent for detection of lipids on thin-layer chromatograms (11). Liposomes may be viewed in primulin solutions with fluorescent microscopy. Aqueous solutions of 0.1 to 0.001 percent primulin are useful. One drop of liposome preparation and one drop of primulin solution are mixed on a microscope slide and viewed with a filter system having an excitation range of 300-400 nm and a barrier filter above 420 nm. Addition of fluorescent microspheres of determined size allows estimation of liposome size by direct comparison.

3. Use of CHAPS Detergent for Liposome Preparation

β-Octyl glucoside and other detergents have been used to solubilize lipids before dialysis to remove the detergent and form liposomes (7, 8, 9). The synthesis of CHAPS detergent was reported in the literature in 1980 (3) and then became available commercially. When 60 mM CHAPS and β-octyl glucoside are compared by dissolving DPPC (7.9 mg in 5 ml) in each detergent in 50 mM KCl and dialyzing against 50 mM KCl at a temperature (41°C) above the transition point of DPPC, the liposomes produced are in greater numbers with greater uniformity in the case of CHAPS. We conclude that the use of CHAPS detergent is preferable to β-octyl glucoside, at least when synthetic DPPC is the phospholipid of choice.

Discussion

We have prepared these liposomes for the purpose of delivering water-soluble materials to the food vacuoles of paramecia and for the incorporation of fluorescent lipids into the vacuolar membranes of paramecia. The size of these liposomes is such that they are rapidly engulfed by *Paramecium multi-micronucleatum* and *Paramecium tetraaurelia*. The paramecia containing fluorescent and non-fluorescent liposomes in their food vacuoles may be photographed successfully with Kodak 400 Daylight film. Further experiments with uptake of liposomes by paramecia are in progress.

Summary

We make three points about liposome preparation:

1. Liposomes, including fluorescent liposomes, may be prepared through reverse-phase evaporation by autoclaving.
2. Liposomes prepared with synthetic DPPC may be visualized with primulin, a water-soluble dye with an affinity for lipids, and fluorescence microscopy.
3. CHAPS detergent is superior to β-octyl glucoside detergent in liposome

preparation by the detergent dialysis method with synthetic DPPC. Additionally, fluorescent and non-fluorescent liposomes prepared by these methods are engulfed rapidly by two species of paramecia.

Acknowledgments

We thank W. N. Doemel for suggesting the use of primulin and N. E. Leal for discussions about these experiments.

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Immunotherapy of the IR162 Rat Myeloma

Kara Eberly and Kelley Kirwan
Department of Biology
Saint Mary's College, Notre Dame, IN 46556

The IR162 transplantable rat myeloma is a spontaneous tumor in a Louvain rat (2, 3). The tumor has been established in cell culture as the Ta4 line, and tumor cells grown *in vitro* and *in vivo* have the morphology of plasmablasts (8, 10, 11). The tumor cells secrete immunoglobulin E *in vitro* and *in vivo*, and express IgE on their cell surfaces (9, 10). The growth and metatasis of the tumor has been well described, and the tumor cells have been extensively examined *in vitro* for their sensitivity to a number of cytotoxic and immunomodulatory agents (8-11). The IR162 tumor was therefore considered a potential model for immunotherapy of multiple myeloma.

Nonspecific immunity has often been shown to be due to natural killer cell activity (6, 14, 18), so nonspecific immunity was examined by the intravenous injection of normal spleen cells into tumor bearing animals, and the induction of lymphokines, which have been reported to enhance natural killer cell activity (18, 20). Specific immunity was evaluated by the use of spleen cells from tumor bearing animals and immunization with live and killed tumor cells.

Materials and Methods

Single injections of normal spleen cells. Young adult Lou/M rats 3 to 4 months of age were injected subcutaneously on the dorsum with 3 million IR162 tumor cells from a disaggregated subcutaneous tumor suspended in 0.5 ml Eagles Minimum Essential Medium (MEM). On day 8 after injection, when no tumors were yet palpable, 5 x 10^7 nucleated spleen cells from normal rats of the same sex and age were suspended in MEM and injected intravenously via the tail vein. Control animals received intravenous diluent alone. Animals were observed twice weekly for the appearance of tumors. Tumor size was measured by calipers and expressed as the product of two tumor diameters.

Multiple injections of normal spleen cells. Eighteen 3 to 4 month old Lou/M rats were injected with tumor cells as above and divided into 3 groups. Group I consisted of the control animals, which were intravenously injected with MEM on day 8. Group II received 5 x 10^7 spleen cells from untreated rats on day 8. Group III received spleen cells from untreated rats on days 1, 8, 15, and 22. Tumor size, expressed as the product of two tumor diameters, was observed at days 28 and 35; and tumor weight was determined at autopsy on day 40. Groups were compared by Student's T test.

Spleen cells from tumor-bearing rats. The protocol was the same as for the single normal spleen cell injections except that Group I (9 rats) received 5 x 10^7 spleen cells from rats which had been subcutaneously inoculated with the tumor on day 0. Group II animals (4 rats) were controls and received an intravenous injection of diluent on day 8. Tumor weights at autopsy were compared using Student's T test.

Lymphokine induction followed the procedure of Ruscetti *et al* (17). Five groups of 5 young adult male Lou/M rats were treated as follows. On day -7 groups A, C, and D received an 0.5 ml tail vein injection of an oil in water emulsion containing 2 mg killed acetone dried *Mycobacterium tuberculosis* kindly donated by Dr. Ruth Neta, University of Notre Dame. Group B received the same vaccine

intraperitoneally, and Group E was untreated. On day 0 all rats were inoculated subcutaneously with 1 million IR162 tumor cells in 0.5 ml MEM. Groups A and B received single 0.5 ml intravenous challenge injections containing 100 mg old tuberculin (Jensen) on day 8. Group D received 5 mg old tuberculin(OT) intravenously on days 1, 4, 8, 11, 15, and 18. Groups C and E were unchallenged. Tumor development was observed as above.

Two approaches were taken to evaluating the development of specific immunity to the IR162 myeloma. First, 3 to 4 month old Lou/M rats were injected subcutaneously with 1 million Ta4 cultured IR162 tumor cells, which had been killed by 20 minute exposure to Mitomycin C (20 μg/ml) and emulsified 1:1 in Freund's Complete Adjuvant. Cultured tumor cells were chosen because they would not contain other cell types found in tumor masses. Rats subsequently received 3 biweekly injections of Ta4 cells alone. For allogeneic controls, Sprague-Dawley rats were immunized with 10 million live tumor cells twice, 3 weeks apart. Previous work (8) had shown that this dosage would not produce tumors in Sprague-Dawley rats, but the initial growth of tumor cells should provide increased stimulation of the immune system. In the second approach rats were given a subcutaneous injection of 5×10^5 tumor cells, which caused tumors in less than half the animals; and tumors were allowed to develop. Then the tumor free rats and another group of rats which had not previously received tumor cells were again given that inoculum and compared for the frequency of tumor development to see if the initial failure to develop tumors resulted in immunity.

Antibody mediated cell cytotoxicity was evaluated by the method of Ohanian (16). Ta4 cells were washed twice, suspended in veronal buffered saline (VBS), which had been supplemented with 0.5 mM $MgCl_2$ and 0.1% gelatin, and diluted to 10^5/ml. A 1/10 dilution of serum (0.1 ml) from immunized or control rats and tumor cells (0.1 ml) were incubated together at 30C for 30 minutes to allow antibody to bind to tumor cells; then cells were washed twice in VBS to remove unbound antibody. A 1/10 dilution of rabbit complement (0.1 ml) was added to the cells and incubated at 37C for 30 minutes. Finally 0.1 ml 0.4% trypan blue dye was added and live and dead tumor cells were counted in a hemocytometer after 4 minutes. Data were reported as percent killed.

Cell mediated cytotoxicity was evaluated by preparing cultured Ta4 tumor cells as above, suspending them at 10^5/ml in RPMI 1640 medium supplemented with 10% fetal calf serum, to which nucleated spleen cells were added at ratios of 2:1, 10:1, and 100:1, and incubated 5 hours at 37C, which is an appropriate time for NK cell killing (6). Spleen cells were disaggregated through a mesh screen, purified on Histopaque (Sigma Chemical), and washed twice in balanced salt solution. After incubation, trypan blue dye was added as above, and live and dead tumor cells were counted. Note that because of the difference in size, tumor cells may be easily differentiated from spleen cells. Data were expressed as percent killed and total remaining tumor cells.

Results

Intravenous injection of normal spleen cells increased the tumor free period and survival of Lou/M rats injected with IR162 tumor cells. In one experiment involving 10 control and 6 experimental animals, spleen cell therapy increased the mean survival from 55 to 74 days, excluding one treated animal which died without evidence of tumor at 280 days. In a second experiment control rats had a mean survival of 70 days, and 1 of 8 remained tumor free when the experiment was terminated at 180 days; experimental animals had a mean survival of 121

days with 4 or 8 tumor free at 180 days. The longer survival of controls in the latter experiment probably reflected a slightly lower viability of the initial tumor cell inoculum. Other experiments have shown that the state of tumor development, controlled by inoculum size and day of treatment, is critical to the success of spleen cell therapy.

Multiple spleen cell injections beginning on day 1 after tumor inoculation significantly ($p < .05$ by Student's T test) reduced tumor weight at autopsy (day 40) to 15 g compared to 18 g with one injection or 35 g for untreated animals. Significant reduction in tumor size was also observed at days 28 and 35 (Figure 1).

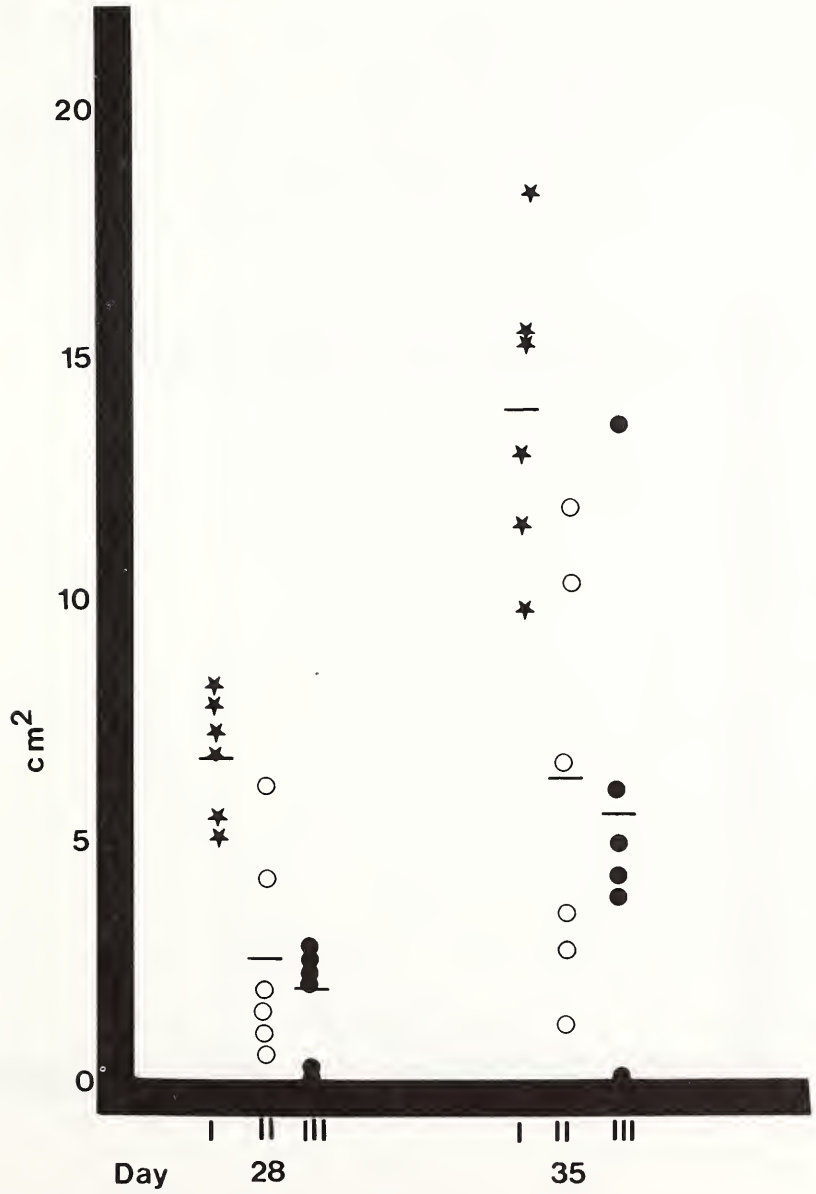

FIGURE 1. *Effect of normal spleen cells on tumor development. (-) indicates the mean value. Tumor size is expressed as the product of two diameters. Group I was untreated, II received a single injection of spleen cells on day 8, III received multiple spleen cell injections. II and III differ significantly from group A ($p < .05$ by Student's T test).*

Lymphokine induction by immunization to *Mycobacterium tuberculosis* and challenge with OT inhibited tumor development (Figure 2). Tumor weight at autopsy showed a similar pattern. The unchallenged groups C and E had mean tumor weights of 18.8 and 14.0 grams respectively. The challenged groups, A and B (100 mg OT intravenously) and D (repeated 5 mg challenges) had mean tumor weights of 10.9, 2.7, and 7.2 grams. Although differences between challenged and unchallenged groups were apparent, they were not significant at the 5 percent level using Student's T test.

Since nonspecific methods proved effective in inhibiting the tumor, and some spleen cell appeared to be responsible, spleen cells from tumor inoculated animals were evaluated as *in vivo* therapy (Figure 3). In contrast to the results with normal spleen cells, cells from tumor bearers significantly enhanced rather than inhibited the tumor growth ($p < .05$ by Student's T test).

When low tumor inoculum was used and tumor appearance in animals which had once failed to develop tumors was compared to animals which had never been exposed to the tumor, there was no apparent or significant difference in the fraction which developed tumors nor in the time for tumor development.

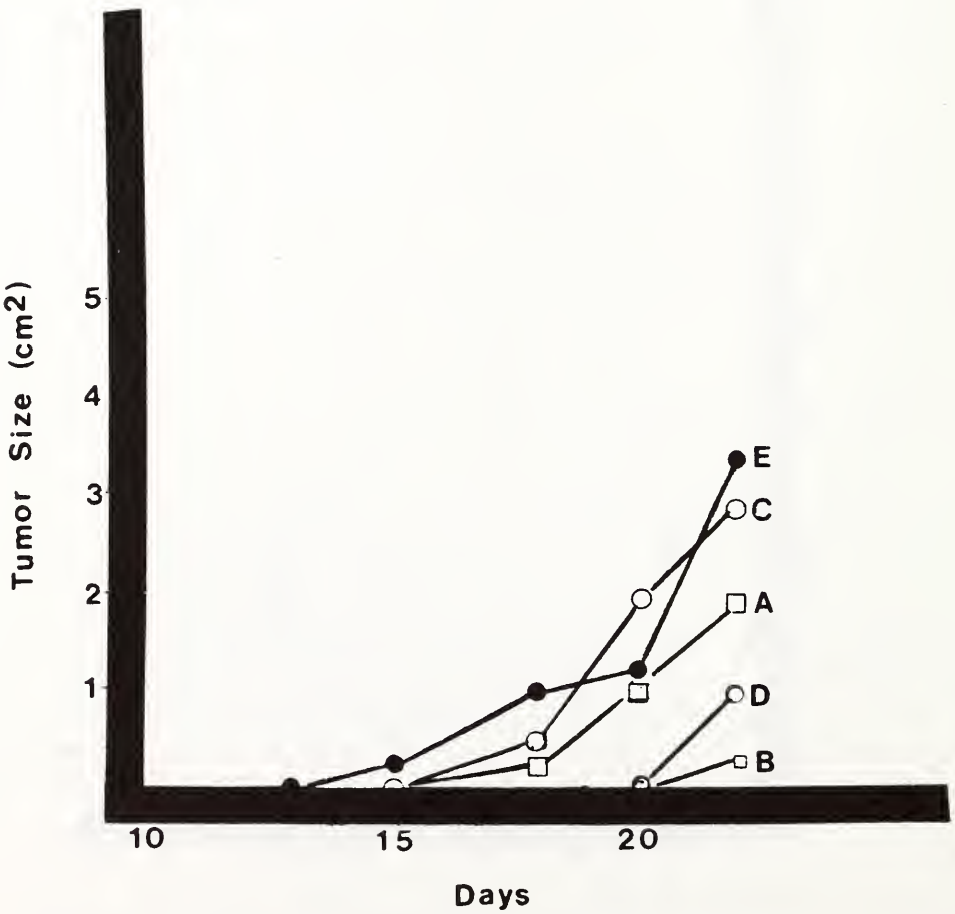

FIGURE 2. *Effect of lymphokine induction on tumor development. Points represent mean tumor size expressed as the product of 2 diameters. Group A—i.v. immunization, i.v. challenge (100mg OT), Group B—i.p. immunization, i.v. challenge (100mg OT), Group C—i.v. immunization, no challenge, Group D—i.v. immunization, repeated low dose (5mg) challenge i.v., Group E—no treatment.*

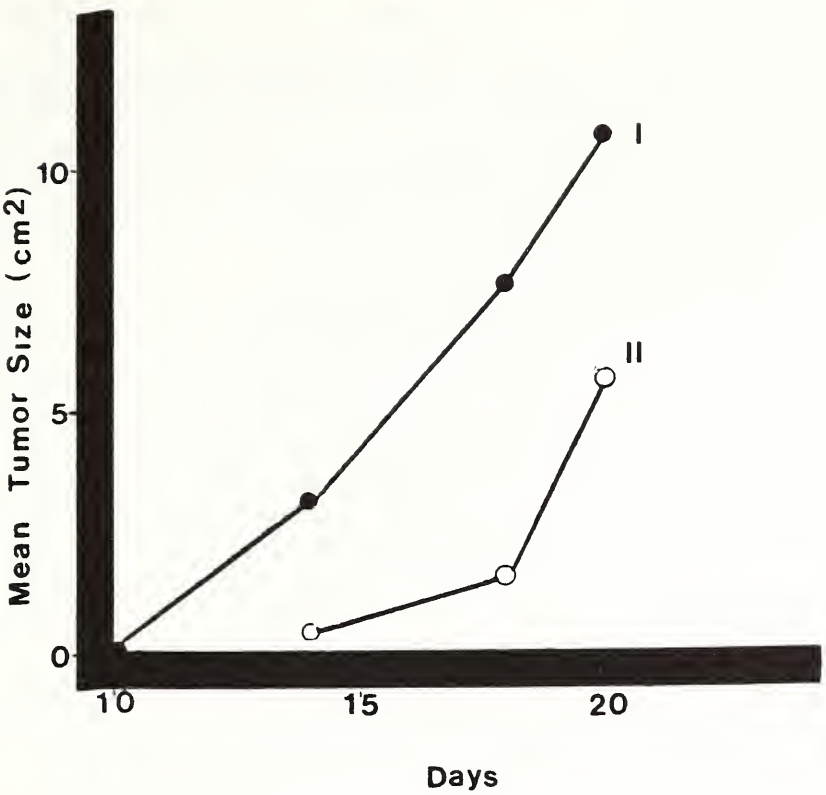

FIGURE 3. *Effect of spleen cells from tumor bearing rats. Group I received spleen cells from rats which were inoculated with a subcutaneous tumor 8 days before. Group II was untreated.*

Rats were immunized with killed tumor cells and evaluated for antibody that could kill tumor cells in the presence of complement. Rabbit anti-rat antibody and rat antibody against the major histocompatability antigens of the Lou/M rat, anti-AgB2, prepared according to the precedure of Shinohara (19), were used as positive controls; normal rat serum was used as a negative control. Rabbit complement was found to give much better results than guinea pig complement with the rat antibody. Data were presented as percent of cells killed, as judged by trypan blue exclusion, and greater than 35% kill was considered positive. No control syngeneic (Lou/M) or allogeneic (Sprague-Dawley) rats produced cytotoxic serum. Two of 5 syngeneic immunized rats had cytotoxic antibody as did 3 or 8 allogeneic rats. When immunized Lou/M rats were killed and serum was tested at various times during the immunization procedure, 1 of 2 sera was positive at 1 week, 2 of 2 at 3 weeks, and 1 of 5 at 6 weeks. Therefore, the development of cytotoxic antibody occurred but was sporadic in immunized syngeneic and allogeneic rats. Sensitivity of the tumor cells to anti-AgB2 antiserum indicated that the tumor cells were susceptible to complement-mediated antibody killing, and that the cells expressed histocompatibility antigens.

Examination of spleen cells from immunized rats above indicated a slightly increased cell mediated cytotoxicity using 100:1 ratio of spleen to tumor cells over that observed with a 2:1 ratio, but there was no significantly increased cell mediated killing in immunized rats. A similar experiment in which rats were killed at two day intervals up to 2 weeks after tumor inoculation, also failed to show enhanced *in vitro* cell mediated killing by spleen cells.

Discussion

Multiple myeloma in humans is a widespread disease which may be treated with cytotoxic drugs but is seldom cured (7). Therefore, there has been interest in immunotherapy of the disease. Mouse plasmacytomas were responsive to specific immunotherapy (1.5.12), but the presence of virus in virtually all mouse cells and the response of the immune system to viruses makes the results somewhat more difficult to interpret.

The rat myeloma cells are virus free as far as is known (4), and the growth and metastasis of the transplantable IR162 myeloma has been thoroughly characterized (2,3,8,11). Therefore, we have attempted to evaluate the responsiveness of this model to nonspecific and specific immune modulation *in vivo*.

The tumor developing at a subcutaneous site was shown to be susceptible to inhibition by intravenously injected normal spleen cells, and multiple injections begun at day 1 were more effective than a single injection at day 8. Tumor size (expressed as the product of 2 diameters) has been shown to increase linearly with time once the tumors become palpable (11). Therefore, although the tumors were not palpable at day 8, it is reasonable to assume that the spleen cells travelled to the subcutaneous site and killed tumor cells, thus reducing the tumor burden. This implies the presence of a cell type in the normal spleen which can kill IR162 cells. The most likely candidate for that cell would be natural killer of NK cell, which is found at high levels in the spleens of rats from 2 to 18 months of age (6). Using our procedure for *in vitro* cell killing, which is admittedly not very sensitive, we were unable to find evidence for cells killing IR162 cells.

However, interferon enhancement of natural killer cell activity has been extensively documented (3,18,20). Since we were primarily interested in the *in vivo* effects of spleen cells, a procedure was chosen which has been shown to induce lymphokines, including interferon, in rats (17). Both single high dose (100mg) and repeated low dose (5mg) challenges with OT inhibited tumor development. Thus lymphokine induction was shown to be effective as nonspecific immunotherapy. Therefore, there is at least suggestive evidence that the cell type responsible for the observed nonspecific immunotherapy of the rat myeloma may be the natural killer cell.

Since the tumor cells were susceptible to antibody mediated killing *in vitro*, they might be susceptible to specific immunotherapy. Several approaches were tried. First, spleen cells from donor rats at the same stage of tumor development as the recipients (day 8) were used. Although tumor cells were found in the spleen later in the course of the disease, tumor cells are not present at this early stage (11). If immunity were developing during tumor growth, concomitant immunity, we might expect to detect increased tumor inhibition. Instead we observed enhanced tumor growth. Attempts to find spleen cells capable of killing tumor cells *in vitro* during the first two weeks of tumor development were also unsuccessful.

We therefore postulate the development of immunosuppressive spleen cells early in myeloma development. Myelomas have been documented to be immunosuppressive (7); but it was somewhat surprising to find the suppression so early in tumor development. Perhaps that helps to explain the spontaneous nature of the rat myelomas and how readily they may be transplanted. Mouse myelomas, in contrast, must be induced by the intraperitoneal injection of pristane (2,6,10,14-tetramethylpentadecane) and generally such an injection is used to "prime" mice for tumor transplantation. This appears to be due to the immunosuppressive activity of the pristane (1). The ability of the rat myeloma to suppress

the immune system early in its development could explain the difference between the two models.

Two methods of tumor immunization were tried. Rats were given a low tumor inoculum so that only about half the animals developed tumors. Then the tumor free rats were compared to rats of the same age which had never been exposed to IR162. The second tumor inoculum was also low. The rats which had been previously exposed to the tumor showed no increased resistance to tumor development; they did not appear to have developed any specific immunity. Other Lou/M rats were immunized with killed cultured tumor cells. Cultured cells were used because they had been cloned and were known to be only IR162 cells (10). Tumors do contain other cell types (15). The sporadic nature of the antibody forming response probably indicates that immunization was not optimum. However, some rats developed antibodies capable of killing the tumor cells. Therefore, the tumor cells do contain antigens capable of eliciting an immune response in syngeneic rats, and those cells are susceptible to antibody-complement mediated killing.

In vitro tests thus demonstrate a potential for the development of protocols for specific immunotherapy of the IR162 rat myeloma, although optimum immunization protocols have not been obtained. However, the IR162 tumor model is definitely susceptible to nonspecific immunotherapy *in vivo*. Since the tumor cells can be used to evaluate drug action *in vitro*, and they themselves are not particularly sensitive to a number of immunomodulataory drugs (9), this provides a promising model for *in vivo* trials of immunomodulators. One could be relatively certain that the action of such drugs was on the immune system rather than directly on the tumor cells. Therefore, results should reflect the response of the host to the drug rather than the sensitivity of individual tumors to those compounds.

Acknowledgments

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CHEMISTRY

Chairman: MARGARET A. CAVANAUGH
Department of Chemistry, Saint Mary's College, Notre Dame, Indiana 46556

Chairman-Elect: ERIC R. JOHNSON
Department of Chemistry, Ball State University, Muncie, Indiana 47306

ABSTRACTS

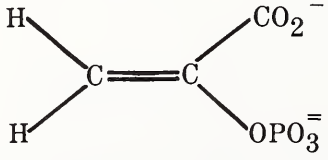
The Effects of Phosphorus Ligand Size on the *cis:trans* Distributions in $W(CO)_4LL'$ Complexes. DANIEL V. BROWN, DENNIS A. DRAKE, CONSTANCE A. KIESLER and JOHN A. MOSBO, Department of Chemistry, Ball State University, Muncie, Indiana 47306.—Thirteen phosphines (L′) have been reacted with $W(CO)_4L$(pyridine) complexes (L = $PPhMe_2$, PPH_2Et or P(*p*-tolyl)$_3$) to produce $W(CO)_4LL'$ products. The *cis:trans* product ratios, measured by integration of 31-phosphorus nmr spectra, generally decreased as the size of L′ increased. Thus, for example, when L = PPh_2Et, the *cis:trans* ratios decreased in the L′ order $PPhMe_2 > PPh_2H > Pet_3 \sim PBu_3 \sim PPh_2Me > PPh_2Et > PPh_2$(iso-Pr) > $PPh_3 \sim$ P(*p*-tolyl)$_3 \sim PPh_2$(tert-butyl). These results are consistent with a rapid basal-apical equilibrium of a square pyramidal $W(CO)_4L$ intermediate.

Computer-Controlled Multiple Standard Addition System for Use with Ion Selective Electrodes. STANLEY L BURDEN and STEPHEN H. FLOWERS. Departments of Chemistry and Computer Science, Taylor University, Upland, Indiana 46989.——An Apple II Plus microcomputer is used to control the delivery of standard solution from a constant rate buret into a solution being analyzed. The computer automatically logs the potential of electrodes immersed in the solution after equilibrium has been reached. The user may specify the number of additions and either the volume of standard to be added for each addition or the magnitude of electrode potential change before the addition is terminated. The user may elect to determine either the electrode slope or the concentration of an unknown solution. In either case the computer computes both the individual results from each addition as well as the average and relative standard deviation of the series. An optional high resolution graphics display of a conventional standard addition plot of the data fit with a least squares line is also available. The system has been tested with both nitrate and chloride ion selective electrodes. Accuracies and precisions comparable to or better than manual standard addition methods are typical but very significant savings in time and analyst effort are realized.

The Isolation and Characterization of a Denaturant-stable Bacterial Protease. NEAL E. COLEMAN and ERIC R. JOHNSON, Department of Chemistry, Ball State University, Muncie, Indiana 47306.——A new protease component that is both stable and active in the presence of 6.0 *M* guanidinium chloride has been isolated from Pronase, a commercially available mixture of proteases isolated from *Streptomyces griseus*. This protease has exhibited esterase activity against N-α-acetyl-L-tyrosine ethyl ester (ATEE), a synthetic substrate for chymotrypsin, both in

the presence and absence of 6.0 *M* guanidinium chloride. This newly isolated protease comprises approximately 10% of the total denaturant-stable ATEE esterase activity found in the Pronase mixture. The unique ion-exchange chromatographic behavior and electrophoretic mobility suggest that this proteolytic enzyme is different than the two denaturant-stable Pronase proteases that have been previously described (W.M. Awad, Jr., *et at.* (1972) *Proc. Nat. Acad. Sci (USA) 69* 2561-2565).

Stereoselective Ligand Interactions with Phosphoenolpyruvate Carboxykinase. THOMAS DUFFY, MYOUNG HEE LEE and THOMAS NOWAK, University of Notre Dame, Notre Dame, Indiana 46556.—— Phosphoenolpyruvate carboxykinase (PEPCK) catalyzes the reversible carboxylation of phosphoenolpyruvate (PEP), with concommitant phosphoryl transfer to a nucleotide acceptor, to form oxaloacetate (OAA) and GTP. Analogues of substrates PEP, GDP and GTP have been used to study



$$\text{(PEP)} + \underset{\text{(IDP)}}{\text{GDP}} + CO_2 \overset{M^{2+}}{\rightleftharpoons} \text{OAA} + \underset{\text{(ITP)}}{\text{GTP}}$$

specific ligand-enzyme interactions with this enzyme. Analogues of the substrate PEP include E and Z-2-phosphoenolbutyrate (E-PEB and Z-PEB), E and Z-3-fluorophosphoenolpyruvate (E-F-PEP and Z-F-PEP), and Z-3-bromophosphoenolpyruvate (Br-PEP). Only Z-F-PEP showed substrate activity (42% V_{max}) with the $M_n{}^{+2}$ activated enzyme. E-F-PEP was a competitive inhibitor (K_I = 23 μM). The PEB diastereomers exhibited stereoselective inhibition (K_I, Z = 32 μM; K_IE = 1.5 μM). PRR titration studies show that $K_I = k_d$ for these ligands and the enzyme-Mn complexes containing Z-PEB and F-PEP resemble the complex containing PEP. Thiol analogues of the nucleotides in which a nonbridge oxygen is substituted by sulfur showed selectivity in interactions. The R and S diastereomers of GTP-αS both demonstrated substrate activity with a V_{max} ratio (R/S) of 0.5 - 1.8. The GDP-α-S pair showed a ratio of 3-5; the ratio was dependent upon the divalent cations used. GTP-β-S exhibited dramatic selectivity with a V_{max} ratio of 0.005 - .01. GDP-β-S showed no substrate activity whereas GTP-γ-S did. The kinetic studies with PEP analogues suggest steric effects are stereoselective at the C-3 position but are not detrimental to ligand binding but perhaps block catalysis. Results with nucleotide analogues indicate the α-P is not very sensitive to events during catalysis, however the β-P where the reaction occurs shows strict selectivity.

Enthalpy of Nucleophilic Addition to Enenitriles. JANE E. HINNERS and TERRY KRUGER, Ball State University, Muncie, Indiana 47306.—— Nucleophilic attack by the 1°, 2°, and 3° amines on cyclohexylidenemalononitrile yields monoadducts. The temperature change accompanying each reaction was recorded in a Parr solution calorimeter. The heat of reaction was calculated. The enthalpies were correlated with the proton affinity of each amine. The structure of the adducts were studied by nuclear magnetic resonance and infrared spectroscopy at different time intervals.

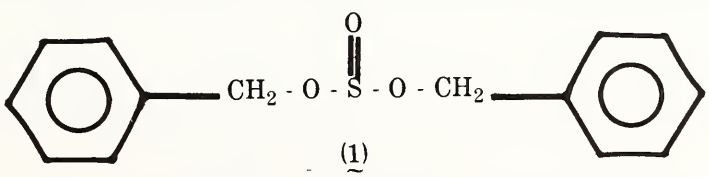
A Spectrophotometric Investigation of the Reaction Between Aqueous Cadmium(II) Ions and Electrolytically Generated Hydroxide Ions. A.J.C.L. HOGARTH, Department of Chemistry, Depauw University, Greencastle, Indiana 46135 and T. S. WEST, Director, Macauley Institute for Soil Research, Craigiebuckler, Aberdeen, Scotland.—— Spectroelectrochemical techniques have made a modest impact on the world of analytical chemistry in the last decade-and-a-half. The products of

research have been thin film cells and sometimes valuable observational data relating to electrochemical reactions. This paper considers a somewhat different approach not using optically transparent cells, but a generally simpler experimental arrangement. Hydroxide ions were generated at an anodically pretreated platinum plate electrode, and their reaction with aqueous cadmium(II) ions observed by attenuations to a collimated beam of radiation passing through the solution and over the electrode surface. The instrumentation and experimental results are discussed briefly.

A 4-H Theory of Photosynthesis by Green Plants. ROBERT H. L. HOWE, Eli Lilly and Company, Lafayette, Indiana 47809.—— A new interpretation of the chemical mechanism of oxygen production by algae is advanced by the author through his many years of investigation, as based on the accepted theory that H_2O is the H-donor during the process of photosynthesis. Therefore, the 4H will be required and 4OH will be released from water. Then, $2H_2O_2$ will be formed and one molecule of O_2 is later generated. The explanation of this interpretation is presented, the peroxide route reaction pathways are discussed, and the significance of the phenomena is related.

The Proteolytic Action of Guanidine-stable Chymoelastase on Low Molecular Weight Peptides. KATHERINE J. JORDAN and ERIC R. JOHNSON, Department of Chemistry, Ball State University, Muncie, Indiana 47306.—— Guanidine-stable chymoelastase, a denaturant-stable proteolytic enzyme produced by the K-1 strain of *Streptomyces griseus*, has been reported to preferentially catalyze the hydrolysis of protein substrates at peptide bonds on the C-terminal side of phenylalanine, tyrosine, and leucine in the absence of denaturant (Y. Narahashi and K. Yoda (1973) J. *Biochem. (Tokyo) 73* 831-841). The results of the current study indicate that, in the presence of denaturant, phenylalanyl and tyrosyl peptide bonds of low molecular weight peptide substrates are hydrolyzed preferentially under the catalytic influence of this enzyme. However, leucyl peptide bonds in three different low molecular weight peptides were not hydrolyzed by this enzyme in the presence of denaturant. Although some denaturant-dependent differences in cleavage specificities are apparent, these observations indicate that guanidine-stable chymoelastase does show cleavage specificity for phenylalanyl and tyrosyl peptide bonds in the presence of 6.0 *M* guanidinium chloride.

Singlet and Triplet Mechanisms in the Photochemistry of Benzyl Sulfite. ROBERT J. OLSEN, BRAD D. MAXWELL and C. ALLEN RIDGEWAY, Department of Chemistry, Wabash College, Crawfordsville, Indiana 47933.—— The photorearrangement and photofragmentation reactions of benzyl sulfite (1) have been examined.



(1)

Direct irradiation in benzene results in the formation of benzyl alcohol, benzyl ether and benzyl phenylmethanesulfonate as the major products. The quantum yield for disappearance of 1 is 0.39 at 254 nm and the reaction is only weakly quenched by isoprene. A crossover experiment indicates that approximately 85% of the benzyl ether is formed by a recombination reaction occurring within a solvent cage.

Acetone sensitized photolysis in benzene at 300 nm affords benzyl alcohol

and bibenzyl as major products. A crossover experiment indicates that the bibenzyl is formed outside the initial solvent cage.

A mechanism to account for these observations will be presented.

Inhibition of ADP-Induced Aggregation of Human Platelet-Rich Plasma Suspensions by the β,γ-Phosphonic Acid Analog of ATP. B. H. RAGATZ, School of Medicine, Indiana University, Center for Medical Education, Fort Wayne, Indiana 46805, P. G. IATRIDIS, School of Medicine, Indiana University, Center for Medical Education, Gary, Indiana 46408 and S. G. IATRIDIS, School of Medicine, University of Athens, Athens, Greece.——Using washed platelet-rich plasma (PRP) suspensions from rabbit, Packham and coworkers found that various naturally occurring nucleoside triphosphates (NTP) inhibited ADP induced aggregation. They also determined that these compounds served as substrates for platelet nucleoside diphosphokinase (NDK). They postulated that NDK usually catalyzed the transfer of membranbound phosphate to ADP as a primary event in aggregation but that various NTP compounds could substitute as phosphate donors and thus block dephosphorylation of the platelet surface.

To test this hypothesis, Born and Foulks incubated rabbit PRP with β,γ-methylene ATP, an ATP analog which cannot be easily hydrolyzed by IM hydrochloric acid or by various phosphatases. They found good inhibition by β,γ-methylene ATP if ADP was added to the PRP 10 seconds later and concluded that labile terminal phosphate is not required on an NTP for ADP-induced aggregation to be inhibited. They suggested that NTP compounds merely bind competitively to the platelet ADP receptor site.

We have expanded the experiments and confirmed the conclusions of Born and Foulks using human PRP suspensions. The following observations were made:

a) The compound, β,γ-methylene ATP, alone showed no intrinsic effects on the light transmitted through a PRP suspension.

b) High doses of β,γ-methylene ATP inhibited the aggregation of PRP exposed to ADP 30 seconds later, but not as extensively as equimolor amounts of ATP.

c) No time dependent changes were observed in the inhibitory profiles when ATP or β,γ-methylene ATP was preincubated with PRP for 5 minutes before ADP addition.

It reamins to be investigated if these observations can be generalized to the platelet ADP receptors of other mammalian species.

Potential Method for the Improved Determination of Lecithin/Sphingomyelin Ratios in Patient Amniotic Fluid Samples. B. H. RAGATZ and B. A. OTFINOSKI, School of Medicine, Indiana University, Center for Medical Education, Fort Wayne, Indiana 46805.——Last year, we reported the optimization of conditions for separation of seven phospholipids by silica gel chromatography and their detection by ten different reagents. Among the seven phospholipids separated, two are of published importance in assessment of fetal lung maturity when extracted from amniotic fluid. These two are lecithin and sphingomyelin.

More recently we have prepared chloroform/methanol extracts of 37 amniotic fluid samples taken from patients at three different hospitals. The concentrated extracts have been spotted on air dried silica gel coated plates, along with lecithin and sphingomyelin standards and developed in $CHCl_3$: methanol: 30% NH_4OH (68:28:4). Resolved lecithin and sphingomyelin have been detected with iodine vapor, 1-anilino-8-naphthalene sulfonate (ANS) (50 mg/100 ml water), rhodamine B (50 mg/100 ml methanol), and 1, 6-diphenyl-hexatriene (DPH) (10 mg/100 ml dichloromethane).

Standard recoveries of lecithin/sphingomyelin mixtures(2:1) using the various detection systems have been calculated and ANS yields the best recovery observed. Using a subpopulation of calculated ratios from one hospital (determined by the Helena Laboratories method), we have found that our results with ANS detection are not statistically different. Data on subsequent deliveries of three infants from this subpopulation indicate that lung development was normal as predicted by both clinical tests.

Effect of the Presence of a *Clostridium sp.* on Fecal Bile Acids of a Gnotobiotic Gerbil. BERNARD S. WOSTMANN AND MARGARET H. BEAVER, Lobund Laboratory, University of Notre Dame, Notre Dame, Indiana 46556.——The gerbil appears to be a better model for the study of cholesterol and bile acid metabolism than the rat, mainly because its bile acid pattern lacks the muricholic acids and their secondary derivatives. Its body cholesterol pools are sensitive to dietary cholesterol. When fed 0.1% of cholesterol with the diet (average U.S. human intake) serum cholesterol will range between 250 and 400 mg/dl.

The germfree (GF) gerbil would appear to be an ideal baseline for the study of microbial effects on sterol metabolism. However, GF gerbils do not reproduce, due to excessive cecal enlargement. Association with a murine-derived hexaflora consisting of *Lactobacillus brevis*, *Streptococcus faecalis*, *Staphylococcus epidermidis*, *Bacteroides fragilis*, var. *vulgatus*, *Enterobacter aerogenes*, and a *Fusibacterium* sp. reduced cecal size and made reproduction possible. This hexaflora will deconjugate most intestinal BAs, but little further secondary modification occurs. Total fecal BA excretion is, surprisingly, substantially higher than in the conventional (CV) gerbil. Upon introduction of a *Clostridium sp.* this pattern changes. Deconjugation is still almost complete but 1/3 of BA is now modified further, resulting largely in keto-acid formation with limited 7α-dehydroxylation. The fecal neutral sterol fraction showed only cholesterol. The CV microflora, on the other hand, largely 7α-dehydroxylates the primary BAs, with limited keto-acid formation. The fecal neutral sterol fraction contained cholesterol and some coprostanol, but no coprostanone.

Total fecal BA excretion of this hepta-flora associated gerbil is now in the CV range. The data indicate that, while in the GF gerbil approximately 1/3 of biliary BA is chenodeoxycholic acid and 2/3 cholic acid, the relative proportion of cholesterol catabolized via chenodeoxycholic acid declines with increasing complexity of the microflora. The addition of the Clostridium sp. thus leads to substantial keto-acid formation and "normalization" of the amount of fecal BA excreted. A next step will be to introduce a gerbil-derived *E. coli* in the hope of introducing 7α-dehydroxylating capacity comparable to that of the CV microflora.

(Supported by the Eagles' Max Baer Heart Fund)

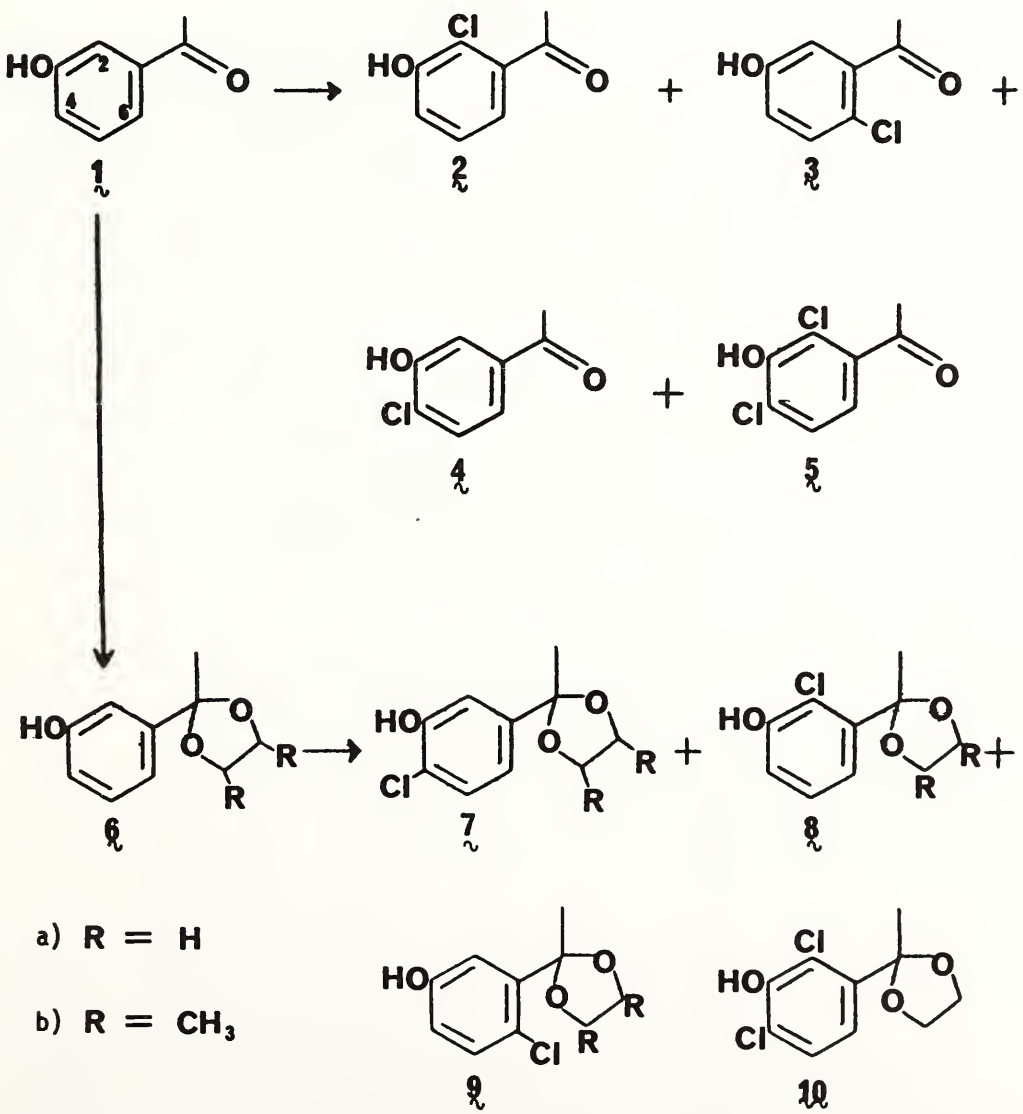
Chlorination of *m*-Hydroxyacetophenone[1]

E. Campaigne, Chung S. Kim and Homer A. Smith, Jr.[2]
Department of Chemistry
Indiana University, Bloomington, Indiana, 47405

Introduction

In connection with another problem (2), we required a quantity of 3-hydroxy-4-chloroacetophenone (compound 4, scheme 1). A search of the literature,

Scheme 1



1. This work was supported in part by contract #N01-HD-0-2818, National Institute of Child Health and Human Development, N.I.H., to Indiana University.

2. National Science Foundation Faculty Fellow, 1976-77, on leave from Hampden-Sydney College, Virginia, 23943.

including formula indices of Beilstein and Chemical Abstracts, indicated that this was apparently a new compound, and a method for its synthesis was needed. In view of the structure of *m*-hydroxyacetophenone (compound 1, scheme 1), and the known propensity of phenol to chlorinate in the *ortho* and *para*-positions, it was assumed that direct chlorination of 1, which is commercially available, with *t*-butyl hypochlorite, which is reported (4) to favor *ortho* chlorination in phenols, would lead to the desired 4-chloro compound 4 as the major product. Indeed, Ginsberg (4) claimed to obtain 2-hydroxy-3-chlorobenzoic acid in 75% yield on chlorination of salicylic acid with *t*-butyl hypochlorite. The presence of the more bulky acetyl group at the *meta*-position in 1 should also favor chlorination at the less hindered 4-position of 1, despite the statement of Harvey and Norman (5) that they had obtained only a 50/50 *ortho/para* ratio on chlorination of phenol in inert solvents with *t*-butyl hypochlorite.

Discussion

Chlorination of *m*-hydroxyacetophenone with *t*-butyl hypochlorite in chloroform solution gave mainly the 2-chloro compound 2 (Table 1, exp. 1) representing substitution at the more hindered position *ortho* to the phenol function, in keeping with Ginsberg's (4) results. It was possible to crystallize some of 2 from the crude mixture, and characterize it as 2-chloro-3-hydroxyacetophenone by spectral data and conversion to the known 2-chloro-3-hydroxybenzoic acid (6). Recrystallized 2 gave a glc peak identical to that of the major component in the product mixture.

We then turned our attention to the ethylene ketal 6a, on the assumption that a more bulky and less polar group at this position should favor chlorination at the 4-position over the 2- and 6-positions favored in the simple ketone. Ketalization of 1 in ethylene glycol using the usual acid catalyst, *p*-toluenesulfonic acid (9) gave considerable trouble, even under nitrogen. Some polymerization occurred, and the reaction reached equilibrium at only about 60% conversion. The maximum yield of vacuum-distilled material was 48%. However, the reaction gave complete conversion using a zinc chloride-phosphoric acid catalyst (8), and pure

TABLE 1. *Chlorination with t-Butyl Hypochlorite*

Expt.	Sub.-strate	Solvent	Temp., C°	Crude product compn., %[a]				
				Starting Material	2/8a	3/9a	4/7a	5/10
1	1	$CHCl_3$	15	13	49	24	6	9
2	6a	THF	0	11	10	33	46	–
3	6a	$CHCl_3$	25	3	31	16	48	–
4	6a	$CHCl_3$	0	4	22	20	54	2
5	6a	$CHCl_3$	-20	4	20	19	56	–
6	6a	C_6H_6-Toluene 9:1	0	5	24	9	62	–
7	6a	C_6H_6-$CHCl_3$, 9:1	0	–	24	11	65	–
8	6a[b]	C_6H_6-$CHCl_3$ 9:1	0	~30	~20	–	~20	~30

[a] Analysis of volatile products by glc using area normalization.
[b] Undistilled ketal 6a was used in this experiment.

6a was obtained in high yield by recrystallization. This avoided the distillation process, which caused decomposition of the ketal, due to the acid character of the phenol.

Indeed, chlorination of 6a gave much more 4-substitution (table 1). Selectivity for the 4-position was better in less polar solvents at lower temperatures. A 9:1 (v:v) mixture of benzene and chloroform at 0° was shown to give 65% of 7a in the crude product mixture (Table 1, exp. 7). Certain samples of 6a, even after repeated recrystallization, gave a much different product mixture, containing substantial amounts of dichloroketal 10 and starting material (Table 1, exp 8), and a reaction rate about ten times faster. The selective monochlorinations listed in experiments 2-7 (Table 1) could only be consistently obtained using samples of ketal that had been distilled from a flask containing potassium carbonate and recrystallized from carbon tetrachloride. We attribute the dichlorination to the presence of traces of base, leading to the formation of phenolates which are much more reactive than unionized phenols (5). In addition, monochloro products 7 and 8, being more acidic, are more easily converted to phenolates, which chlorinate further to 10. Support for the basecatalyzed side reaction was provided by an experiment in which distilled ketal 6a was chlorinated in the presence of a quaternary ammonium hydroxide. Substantial dichlorination was observed. Also, dichlorination was decreased by using undistilled ketal that had been recrystallized from carbon tetrachloride containing acetic acid. However, acid-recrystallized material was not stable on storage.

Selective hydrolysis of a crude chloroketal mixture, prepared as in experiment 7 (Table 1), was carried out under mild conditions. With dilute acetic acid in a water-tetrahydrofuran mixture at reflux for four hours, ketal 7a was completely hydrolyzed, with little or no hydrolysis of 8a or 9a. The latter two ketals, with chlorine *ortho* to the ketal-containing side chain, may be stabilized by the steric or electronic effects of the chlorine atom. Following selective hydrolysis, pure ketone 4 was crystallized, and its structure proven by conversion to the known 4-chloro-3-methoxybenzoic acid (3). Augmentation of the glc peak assigned to 4 with an authentic sample confirmed the analytical assignments (Table 1).

Keton 4 was prepared much more cleanly, and in better yield, from ketal 7b. Chlorination of ketal 6b, derived from 2,3-butylene glycol, with *t*-butyl hypochlorite still gave a mixture of products having about the same distribution among the isomers 7b (65%) 8b and 9b. Thus the more bulky 2,4,5-trimethyl-1,3-dioxolane group did not alter the chlorination ratios appreciably. However, this approach offered the advantage of isolating the pure 4-chloro derivative 7b. While hydrolysis of this compound was more difficult, it was not necessary to use care to avoid hydrolysis of unwanted isomers, as was the case with hydrolysis of 7a. Thus recrystallized 7b, obtained in about 60% yield, was converted to 4 in nearly quantitative yield.

The structural assignments of ketones 2 and 4 were further confirmed by a study of dichlorination products. Treatment of ketone 1 with two equivalents of *t*-butyl hypochlorite resulted initially in formation of 2, as shown by glc. As this peak waned, the peak assigned to 2,4-dichloro-3-hydroxyacetophenone (5) increased. Dichloroketone 5 was isolated as its 4-nitrophenylhydrazone, which had a doublet of doublets, $j = 8$ Hz, in the pmr assigned to vicinal hydrogens on the phenol ring. Thus compound 5 was either the 2,4- or 2,6-dichloroketone. However, the identical *p*-nitrophenylhydrazone was obtained by dichlorination of ketal 6a, followed by hydrolysis. Since ketal 6a is known to give mainly the 4-chloro derivative 7a on monochlorination, dichlorination must produce 10, the 2,4-dichloro derivative. Therefore compound 2 must have a chlorine at position 2.

Experimental

Melting points were determined on a "uni-melt" Thomas Hoover capillary melting point apparatus and are uncorrected. Infrared spectra were obtained on a Perkin-Elmer Model 137-B infrared spectrometer. Nuclear magnetic resonance spectra were obtained on a Varian T-60A spectrometer and in certain instances on a Varian 220 MHz spectrometer. Mass spectra were obtained on a Hewlett-Packard Model 5992A GC/MS mass spectrometer or on a Varian CH7 mass spectrometer. Gas chromatograph analysis was accomplished using a Varian Model 3700 gas chromatograph equipped with a flame ionization detector, with a 3 ft. x 1/8 in. column packed with 5% Carbowax 20 M on Gas Chrom Q, initial temp 150°, increased on 16°/min to 220°. Elemental analyses were performed by Midwest Microlab, Indianapolis.

Chlorination of 3-Hydroxyacetophenone.

A solution of 2.72 g (20 mmole) of 3-hydroxyacetophenone (1)[3], dissolved in 20 mL of chloroform, was cooled to about 15°, and 2.17 g (20 mmole) of *t*-butyl hypochlorite (7) was added dropwise in subdued light, with cooling to maintain temperature at about 15°C. The mixture was allowed to stand overnight in the dark. Concentration on the rotary evaporator gave an oily residue, which was analyzed by glc, using area normalization. It contained 13% of 1, 49% of 2, 24% of 3, 6% of 4 and 9% of 5. When the residue was cooled overnight, part of it crystallized to give 1 g of material, melting at 61-63°, that gave a glc peak identical to the major component of the crude product. It was identified as 2-chloro-3-hydroxyacetophenone (2) in the following way.

Ketone 2 had the following properties: ir 3225 (OH), 1665 cm^{-1}(CO); pmr (acetone-d_6) δ 7.4-6.8 (m, large single peak at 7.10, 3H, ArH), 2.58 (s, 3H, CH_3).

2-Chloro-3-hydroxyacetophenone 4-Nitrophenylhydrazone.

A small sample of 2 was converted to the 4-nitrophenylhydrazone, (10) m.p. 188-189° after two recrystallizations from alcohol; pmr (acetone-d_6) δ 9.40 (s, 1H, NH), 8.17 (d,2H, J = 9 Hz), 7.13 (d, 2H, J = 9 Hz, ArH), 7.2-6.7 (m, 3H, ArH), 3.6-2.4 (br, 1H, OH, D_2O exchangeable), 2.37 (s, 3H, CH_3).

Anal. calc'd for $C_{14}H_{12}N_3O_3Cl$: C, 55.08; H, 3.93; N, 13.77. Found: C, 55.11; H, 4.12; N, 13.87.

2-Chloro-3-hydroxybenzoic Acid.

Oxidation of ketone 2 with sodium hypochlorite solution gave a crystalline acid, recrystallized from toluene-hexane mixture, and melting at 164-166°. Beyer (1) reported m.p. 158-159° for this acid, 219-220° for 4-chloro-3-hydroxybenzoic acid, and 178-179° for 6-chloro-3-hydroxybenzoic acid. However, Hodgsen and Rosenberg (6) give 161.5° as m.p. of 2-chloro-3-hydroxybenzoic acid.

2-(3'-Hydroxyphenyl)-2-methyl-1,3-dioxolane (6a).

A. *Use of Toluenesulfonic Acid.* A mixture of 150 g (1.1 mole) of 1, 70 g of ethylene glycol, 2.3 g of *p*-toluenesulfonic acid monohydrate, and 1.5 liter of benzene was refluxed until no further water collected in the Dean-Stark trap

3. Technical grade *m*-hydroxyacetophenone, m.p. 86-88° (Aldrich) was recrystallized from toluene or hot water to melting point 94-96°. However, the hot water recrystallization, using activated carbon, gave purer product with less tars carried along.

(100 h). The acid was neutralized with 2.3 g sodium methoxide, the warm mixture filtered through a fluted filter, washed with brine, and concentrated on a rotary evaporator to give 139 g (70%) of pale yellow crystals, m.p. 83-85°. The crude product, mixed with 0.7 g potassium carbonate, was distilled at 1 torr using a short column and air-cooled condenser, to give a colorless oil, b.p. 130-140°, which solidified. Recrystallization from carbon tetrachloride gave 93 g (47%) of white crystals of 6a, m.p. 90-91°; ir (KBr) 3250 (OH), 1140-1070 (ketal CO)cm^{-1}, pmr (DMSO-d_6) δ 9.1 (1H, OH), 7.3-6.5 (m, 4H, ArH), 4.2-3.6 (m, 4H, -CH_2-), 1.65 (s, 3H, CH_3).

Anal. calc'd for $C_{10}H_{12}O_3$: C, 66.65; H, 6.71; M.W. 180. Found: C, 66.48; H, 6.54; M^+ 180.

B. *Use of Phosphoric Acid/Zinc Chloride.* In a 500 mL 3-neck flask, equipped with magnetic stirrer, nitrogen inlet and Dean-Stark trap was placed 13.6 g (0.1 mole) of 1, 31 g (0.5 mole) of ethylene glycol, 0.2 g of anh. zinc chloride, 0.2 mL of 85% phosphoric acid and 70 mL of toluene. The mixture was refluxed until tlc showed no ketone present. The mixture was cooled, 100 mL of ether added, and the solution washed with aqueous bicarbonate solution. The aqueous layer was washed once with ether, and the combined ether extracts dried ($MgSO_4$) and concentrated (rotary evaporator) to give 16.1 g (89%) of white crystalline 6a, melting at 87-91°. Recrystallization from cyclohexane gave white crystals melting at 89-91°, identical in ir spectrum to 6a prepared above.

3-Hydroxy-4-chloroacetophenone (4).

A. *By Hydrolysis of 7a.* In a three-neck flask with drying tube and mechanical stirrer, a solution of 15 g (83 mmol) of 6a in 270 mL of benzene and 30 mL of chloroform was cooled in an ice-bath. In dim light, a mixture of 9.49 g (87 mmoles) of *t*-butyl hypochlorite in 30 mL of 9/1 benzene/chloroform was added dropwise. After 4 h, the mixture was allowed to stand at room temperature overnight. Analysis by glc showed a mixture containing approximately 65% of 7a, with 24% of 8a and 11% of 9a. The oily residue, obtained on removal of solvents under vacuum, was refluxed for 3 h in a heterogeneous mixture of 250 mL THF, 250 mL water and 25 mL glacial acetic acid. The aqueous solution was extracted with ether, and the ether extracts combined, washed with brine, saturated sodium bicarbonate, water, and then dried ($MgSO_4$). Concentration gave a semi-solid residue that recrystallized from toluene to give 7.3 g (48%) of 4, m.p. 101-103°. Several recrystallizations from toluene gave an analytical sample, m.p. 106-107°; ir 3290, 1660 cm,s^{-1}; pmr (acetone-d_6) δ 2.54 (s, 3H, CH_3), 7.46 (s, 2H, ArH), 7.58 (s, 1H, ArH), 7.5-8.5 (broad, 1H, OH).

Anal. Calc'd for $C_8H_7O_2Cl$: C, 56.33; H, 4.14; M.W., 170. Found: C, 56.15; H, 4.21; M^+, 170.

The identity of the 4 peak in the glc of the chlorination mixture of 1 was established by augmenting this peak with an authentic sample of 3-hydroxy-4-chloroacetophenone, prepared as described.

B. *By Hydrolysis of 7b.* A solution of 11.5 g (0.047 mole) of 7b (see below) in a mixture of 100 mL of THF, 80 mL of water and 20 mL of conc. HCl was refluxed for 2 hrs. The solution was then concentrated on a rotovap, extracted three times with ether, and the combined ether extracts washed with water, then saturated sodium bicarbonate solution, and finally with brine. After drying over anhydrous $MgSO_4$, concentration of the ether gave 7.6 g (94%) of 3-hydroxy-4-chloroacetophenone (4), melting at 101-104°.

3-Hydroxy-4-chloroacetophenone 4-Nitrophenylhydrazone.

A sample of 4 was converted to the 4-nitrophenylhydrazone, m.p. 231-232°.

Anal. calc'd for $C_{14}H_{12}N_3O_3Cl$: C, 55.08; H, 3.93; N, 13.77. Found: C, 54.80; H, 4.07; N, 13.58.

3-Methoxy-4-chlorobenzoic Acid.

Treatment of 4 with methyl sulfate and potassium carbonate in acetone, followed by oxidation with sodium hypochlorite solution, gave 3-methoxy-4-chlorobenzoic acid, m.p. 212-214°. Gibson (3) gives m.p. 211° for this acid, 160° for 3-methoxy-2-chlorobenzoic acid and 173° for 3-methoxy-6-chlorobenzoic acid.

2,4-Dichloro-3-hydroxyacetophenone 4-Nitrophenylhydrazone.

An authentic sample of 5 was prepared by treating 3.0 g (16 mmol) of ketal 6a in 50 mL benzene and 6 mL of chloroform with 3.9 g (35 mmols) of *t*-butyl hypochlorite in 9 mL of benzene and 1 mL of chloroform, added dropwise, with cooling. After 4 hrs., the mixture was allowed to come to room temp and let stand overnight. Total hydrolysis of this mixture with dilute hydrochloric acid and THF gave an oil, the main constituent of which had a retention time identical to the peak assigned structure 5.

Chlorination of 1 with two equivalents of *t*-butyl hypochlorite, as described above, gave essentially one product, an oil which also had the identical glc retention time as 5. Treatment of this oil with 4-nitrophenylhydrazine gave the 4-nitrophenylhydrazone of 2,4-dichloro-3-hydroxyacetophenone, melting at 245-246° after recrystallization from ethanol-water: pmr (acetone-d_6) δ 8.11, 7.31 (dd, J = 9 Hz, 4H, Ar'H-2',3',5',6', 7.39, 7.04 (dd, J = 8 Hz, 2H, Arh-5,6), 2.34 (s, 3H, CH_3).

Anal. calc'd for $C_{14}H_{11}O_3N_3Cl_2$; C, 49.43; H, 3.26; N, 12.35; M.W. 339. Found: C, 49.27; H, 3.30, N, 12.11; M^+, 339.

2-(3'-Hydroxyphenyl)-2,4,5-trimethyl-1,3-dioxolane (6b).

A mixture of 13.6 g (0.1 mole) of 1, 12.5 g (0.15 mole) of technical grade 2,3-butanediol (a mixture of stereoisomers), 0.2 g of anh. zinc chloride, 0.2 mL of 85% phosphoric acid and 70 mL of toluene was refluxed till all ketone had disappeared (by tcl). After cooling, 100 mL of ether was added, and the mixture worked up as above, to give 18.3 g (88%) of white crystals. Twice recrystallized from carbon tetrachloride, the material melted at 100-103°; pmr (acetone-d_6) δ 1.1 (d, 3H, CH_3), 1.2 (d, 3H,CH_3), 1.53 (s, 3H, CH_3), 3.3-4.3 (m, 2H, =CH-), 6.6-7.3 (m, 4H, ArH), 8.2 (s, 1H, OH).

This compound showed only one spot on tlc. The broad melting point is due to the presence of stereoisomers, as it is a mixture of 2 *meso* and one *dl*-pair of compounds.

Anal. calc'd for $C_{12}H_{16}O_3$; C, 69.23; H, 7.69; M.W. 208. Found: C 69.13; H, 7.84; M^+, 208.

2-(4'-Chloro-3'-hydroxyphenyl)-2,4,5-trimethyl-1,3-dioxolane (7b).

A solution of 6b (20.8 g, 0.1 mole) in 400 mL of a 9:1 benzene/chloroform mixture was cooled to 0°, and 12.3 g (0.11 mole) of t-butyl hypochlorite added dropwise with vigorous stirring. Addition of the hypochlorite required about 1.5-2.0 hrs, after which stirring at 0° was continued for 2 hrs, then continued while the reaction mixture was allowed to come to room temperature overnight. Solvents were removed on a rotovap, and the residual oil was triturated with petroleum

ether, and stored in the cold overnight. The white solid was collected and washed with cold petroleum ether, to give 14.6 g (60%) of 7b, melting at 89-93°. An analytical sample, recrystallized from carbon tetrachloride/petroleum ether melted at 92.5-95°: pmr ($CDCl_3$); δ 1.14 (d, 3H, CH_3), 1.26 (d, 3H, CH_3), 1.6 (s, 3H, CH_3), 3.73 (m, 2H, CH), 5.82 (s, 1H, OH), 7.01 (dd, 1H, ArH-6'), 7.21 (d, 1H, ArH-2'), 7.25 (d, 1H, ArH-5').

Anl. calc'd for $C_{12}H_{15}ClO_3$: C, 59.39; H, 6.19; Cl; 14.62. Found: C, 59.33; H, 6.26; Cl, 14.64.

Summary

3-Hydroxy-4-chloroacetophenone has been prepared from m-hydroxyacetophenone, by chlorination of its ketal derived from 2,3-butylene glycol, using t-butyl hypochlorite. Direct chlorination of m-hydroxyacetophenone gave a mixture, in which 2-chloro-3-hydroxyacetophenone predominated, with some 6-chloro-3-hydroxyacetophenone, and only a little of the desired 4-chloro isomer. 4-Chloro-3-hydroxyacetophenone and 2-chloro-3-hydroxyacetophenone have been isolated and characterized, and their structures confirmed by spectral data, and conversion to known derivatives. 2,4-Dichloro-3-hydroxyacetophenone was isolated as its p-nitrophenylhydrazone, and its structure confirmed by spectral data and alternate synthetic pathways.

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An Interactive Program for the Interpretation of Patterson Functions and Its Uses

TANYA L. CURTIS, P. MICHAEL HAFFLEY and J. C. HUFFMAN
Molecular Structure Center, Department of Chemistry, Indiana University,
Bloomington, Indiana 47405

Introduction

The fact that the phase of F_{hkl}[1] cannot be observed experimentally is a prominent restriction in crystal-structure analysis. A. L. Patterson approached this problem and determined that although a portion of the data is missing for a complete reconstruction of a picture of the crystal structure by Fourier methods and the F_{hkl}'s are not fully available, that the squares of the F_{hkl}'s, which do not involve their phases, are completely available. Patterson solved the problem of deriving information from these $|F_{hkl}|$'s by applying the "convolution" function of Fourier theory (4,5). Thus he derived a function in the form of a Fourier series depending on the $|F_{hkl}|$'s alone, now referred to as the Patterson function (1).

While automatic Patterson solving programs have been developed, many crystallographers prefer to interpret the Patterson function "by hand". A FORTRAN program, PATTI (PATTerson Interpreter), has been written to interactively manipulate the vectors and thus solve molecular structures from the Patterson peaks. While most of the functions carried out by PATTI are trivial in nature, the program provides a convenient mechanism for keeping track of the various vectors and trial positions. PATTI allows the user to generate trial coordinates using combinations of the Harker lines and planes and to examine these trial positions for other interactions. Subroutines are available which also allow the addition or subtraction of suspected interatomic vectors from trial positions already determined. A block diagram of PATTI is shown in Figure 1. The pro-

Subroutine name	Function
PLIST	Generates worksheet indicating possible Harker Vectors and lists various transfomations of X Y Z
HARKER	Calculates Harker symmetry operators and identifies peaks lying on Harker lines and planes
BUILD	Uses Harker planes and lines to define atomic positions (Creates test atom array)
READ20	Reads existing peaks from external tape into test atom array
WRITE20	Write test atom array onto external tape
DELXYZ	Add or subtract known vectors to existing atoms in array
LISTP	List contents of test atom array
XLIST	List Patterson peaks
DELTA	Look for vectors between test atoms
HALFX	Look for 2x 2y 2z peaks (centrosymmetric structures)
CHANGE	Manipulate coordinates in test atom array

FIGURE 1. *Block diagram showing the major subroutines and their functions in program PATTI.*

gram, written in standard FORTRAN, has been implemented on a CDC CYBER172 computer operating with the KRONOS operating system.[2]

Use of the Program

To fully test the program, a data set was obtained on a crystal of a 2:1 mixture of bis-triphenylphosphine zinc(II)bromide and triphenylphosphinetetrahydrofuran zinc(II)bromide. This complex was considered an excellent test of the potential of the program due to the presence of several heavy atoms. The compound crystallizes in space group C2/c with cell dimensions (at -160° C) of a = 35.433(20), b = 16.330(7), c = 15.391(7)Å, β = 108.70(2)°, and D_{calc} = 1.585 gm cm^{-3}. Data were collected at a low temperature (-160°C) using automated diffractometry. The diffractometer and data reduction techniques have been described previously (3).

A listing of the 25 largest peaks in the Patterson map as well as all probable Harker lines and planes through peak 50, the origin peak, are given in Table 1. There were three possible points on the Harker line at 0,Y,1/2 and eight points on the Harker plane at X,0,Z, yielding 24 trial atoms for the structure. Subroutine DELTA correctly located four trial atoms for which there were numerous interactions. A full-matrix least squares assigning bromine scattering factors to

TABLE 1. *Analysis of Patterson Peaks for Trial Structure.*

Peak Number	I	Patterson Coordinates x	y	z		Atomic Interactions*	
1	51.6	.0456	0.000	.1451	H	Zn22-Br24	
2	50.0	.0627	.5000	.3398	H	Zn22-Br24	
3	49.4	0.0000	.0761	.5000	H	Zn22-Zn22	Br24-Br24
4	39.8	.1129	0.000	.1551	H	Br2 -Br2	
5	35.9	.2422	.3576	.3981		Zn1 -Zn22	
6	34.9	.2496	.4199	.3309		Br2 -Br24	
7	33.8	.0716	.3839	.2419		Br23-Br24	
8	33.8	0.0000	.2975	.5000	H	Br23-Br23	
9	33.3	-.0730	.3208	.2530		Br23-Br24	
10	33.3	-.0100	.0986	.0988		Zn22-Br23	
11	33.0	.4619	.3186	.2557		Br23-Br24	
12	32.1	-.0219	.3938	-.0924		Zn22-Br23	
13	31.9	.2491	.1413	.1695		Br2 -Br24	
14	31.7	.1751	.4574	.4110		Br2 -Br23	
15	31.2	-.0337	.1812	.2570		Br23-Br24	
16	31.2	.2140	.0455	.4204		Br2 -Br23	
17	31.0	.2323	.4546	.4991		Zn1 -Br23	
18	31.0	.4642	0.000	-.0030	H	Br23-Br23	
19	30.9	.1812	.4559	.4158		Br2 -Br23	
20	30.7	.0405	-.1076	.2455		Br23-Br24	
21	30.4	.0112	.1938	.4048		Zn22-Br23	
22	30.0	.1403	.4233	.1722		Br2 -Br24	
23	29.8	-.0445	-.0771	.3513		Zn22-Br24	
24	29.7	-.0232	.3058	.3984		Zn22-Br23	
25	29.4	.1756	.2416	-.0820		Br2 -Br23	
31	27.3	.3914	0.0000	.5085	H	Br24-Br24	
35	27.0	.4930	0.0000	.3512	H		
46	23.3	.0163	.5000	.1986	H	Zn22-Zn22	
47	23.1	.0014	0.0000	.2600	H		
48	22.8	0.0000	.2185	.5000	H	Br2 -Br2	
50	18.8	0.0000	0.0000	0.0000			

*H denotes a peak lying on a Harker plane or line.

these four atoms converged to R(F) = 0.35, and a difference Fourier yielded the positions of the missing zinc atom, which lies on a crystallographic two-fold axis, and the phosphorous atoms. Subsequent Fouriers were utilized to locate the remaining non-hydrogen atoms in the two molecules, as well as one tetrahydrofuran (thf) molecule of solvation. Full-matrix refinement converged to final residuals of R(F) = 0.122 and Rw(F) = 0.090 for the 2079 observed ($I > 3\sigma(I)$) data (out of 3958 unique).

The final coordinates were input to PATTI for verification of the Patterson map, and the interactions are noted in Table 1. As can be seen, the top 25 peaks are all due (in part) to interactions between the five heavy atoms (two Zn and three Br). There are few interactions due to Zn1, since it lies in a special position in the cell and thus has half weight in the Patterson.

The two different molecules present in the crystal are shown in Figures 2 and 3, and the fractional coordinates are given in Table 2. Complete crystallographic details for the structure are available.[3]

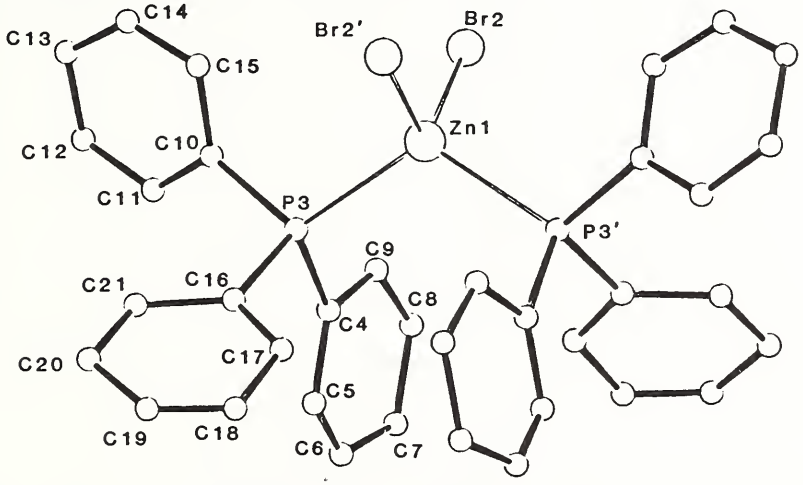

FIGURE 2. *Drawing of the $ZnBr_2(PPh_3)_2$ molecule. Important molecular dimensions are Zn-Br* = 2.378(6)Å; Zn-P = 2.417(13)Å; Br-Zn-Br = 113.5(4)°; P-Zn-P = 114.8(6)° and Br-Zn-P = 104.3(3)°.

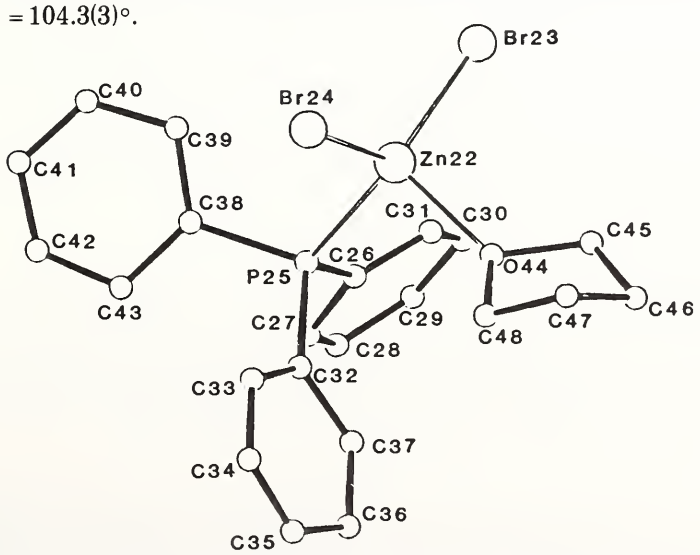

FIGURE 3. *Drawing of the $ZnBr_2(PPh_3)(thf)$ molecule. Important molecular dimensions are Zn-Br* = 2.359(7)Å (avg); Zn-P = 2.436(12)Å; Zn-O = 2.03(3)Å; Br-Zn-Br = 118.1(3)°; Br-Zn-P = 113.5° (avg); Br-Zn-P = 105.9°(avg) and P-Zn-O = 96.6(7)°.

TABLE 2. *Fractional Coordinates for $ZnBr_2(PPh_3)_2.2ZnBr_2(PPh_3)$ (thf).2thf.*

Atom	10^4x	10^4y	10^4z	Atom	10^4x	10^4y	10^4z
Zn1	5000	1902(4)*	7500	C28	5802(11)	722(25)	1807(28)
Br2	4442(1)	1103(3)	6656(3)	C29	5870(19)	1070(40)	1057(43)
P3	5164(3)	2699(8)	6347(8)	C30	6254(13)	1160(29)	1010(31)
C4	4749(10)	3456(22)	5740(23)	C31	6550(10)	918(22)	1745(24)
C5	4846(12)	4286(26)	5787(28)	C32	6865(11)	- 743(24)	3848(26)
C6	4543(15)	4766(32)	5379(34)	C33	7070(14)	-1074(30)	4653(33)
C7	4145(15)	4517(33)	5080(34)	C34	7065(11)	-1895(24)	4888(25)
C8	4047(16)	3636(36)	5042(37)	C35	6836(11)	-2411(25)	4311(27)
C9	4369(14)	3119(29)	5406(31)	C36	6608(11)	-2137(24)	3411(26)
C10	5181(11)	2041(24)	5384(26)	C37	6641(11)	-1304(25)	3206(27)
C11	5121(12)	2361(26)	4539(28)	C38	6832(12)	901(26)	4475(28)
C12	5164(15)	1910(34)	3819(36)	C39	6987(10)	1702(22)	4597(25)
C13	5332(12)	1115(28)	4030(30)	C40	6904(15)	2164(33)	5278(35)
C14	5368(14)	830(30)	4840(33)	C41	7642(13)	- 653(29)	1893(32)
C15	5328(14)	1222(30)	5580(33)	C42	6513(15)	1126(36)	5565(37)
C16	5625(10)	3300(22)	6693(24)	C43	6563(11)	605(24)	4918(26)
C17	5706(11)	3737(24)	7455(26)	O44	7579(7)	- 616(15)	2798(16)
C18	6077(12)	4243(25)	7774(28)	C45	6656(13)	1886(30)	5727(30)
C19	6302(13)	4239(29)	7260(32)	C46	7824(12)	-1482(29)	1886(30)
C20	6249(13)	3804(29)	6446(32)	C47	8056(11)	-1673(25)	2907(27)
C21	5915(10)	3335(22)	6211(23)	C48	7739(15)	-1423(32)	3260(34)
Zn22	7570(1)	449(3)	3467(3)	C49	4213(21)	3479(45)	2871(48)
Br23	7669(1)	1491(3)	2510(3)	C50	3949(21)	2868(45)	2028(50)
Br24	8047(1)	302(3)	4943(3)	C51	4047(17)	3271(34)	1228(38)
P25	6901(3)	295(7)	3560(7)	C52	4394(17)	3643(34)	1620(41)
C26	6506(11)	579(23)	2530(26)	C53	4462(21)	3728(45)	2431(53)
C27	6118(12)	477(27)	2534(29)				

*Numbers in parentheses refer to the estimated standard deviation of the least significant digit.

Results Using Other Structures

To verify the operation of PATTI, fifteen heavy atom structures of various complexity and symmetry were analyzed using the program. In only one case was the program unsuccessful, due primarily to non-crystallographic symmetry in the cell. After solution using other techniques, the correct atoms were found to indeed correspond to the Patterson peaks.

In one of the structures(2), $C_6H_6RuB_7C_2H_{11}$, the program was able to correctly locate not only the ruthenium atom, but six carbon and four boron atoms.

Acknowledgments

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Notes

1. The expression F_{hlk} represents the structure factor for the plane hkl, and is related to the observed intensity (I_{hkl}) by the expression $F_{hkl} = kI_{hkl}^{1/2}$, where k is a scale factor.
2. A FORTRAN listing of the program is available from the Molecular Structure Center at Indiana University. Address requests to JCH.

3. Complete crystallographic details for the zinc complex are available in microfiche from the Chemistry Library, Indiana University, Bloomington, Indiana, 47405. Request Molecular Structure Center Report No. 82805.

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Initial Investigations Concerning the Redox Properties of an Iron (III)-Chloro Complex

A. J. C. L. Hogarth and Melvin E. Lee
Department of Chemistry, DePauw University, Greencastle, Indiana 46135

Introduction

The Nernst equation always presents difficulties to students because, initially, they are expected to take it on trust; this leads to a certain number of questions regarding its origin, and its derivation may be inappropriate for the course. Ultimately the student may just memorize examples of all the types of calculations for which the equation is used, and then forget them after the final examination. To a teacher this is a major inconvenience because the Nernst equation is a stepping stone to many different experimental examples, and it is a waste of time reteaching it.

Experimental work contained herein formed the basis of a short student research project, and was designed to give the student 'bottom-of-the-ladder' experience with a simple redox reagent not readily available commercially, and also examples of the practical application of the Nernst equation. This experimental work involved the student in selection of a suitable synthetic method for the compound; purification procedures; assay of the compound for iron and chloride; stability studies with both the solid and its aqueous solution; and, finally, a determination of its standard reduction potential potentiometrically having selected an appropriate redox titrant.

The compound itself, tetramethylammonium tetrachloroferrate (III), although having been known for some time (4), has been somewhat neglected analytically. It has been used for studies of conductivity and ultraviolet-visible spectroscopy in non-aqueous solvents (4), and little or no attempt has been made to investigate its redox properties.

Theory

The potential difference between a reference electrode and a suitable indicator electrode throughout a potentiometric titration is governed at all points by the equilibrium ratio of the concentrations of the oxidized and reduced species of the couple. These data may be interpreted theoretically using the Nernst equation (i),

$$E = E^{o\prime} - \frac{RT}{nF} \ln K \quad \ldots\ldots \text{(i)}$$

where E is the measured potential; E°' is a combination of the standard potential of the reference electrode used and the standard potential of the redox couple; R is the universal gas constant; T is the absolute temperature; n is the number of electrons transferred; F is the Faraday; and K is the equilibrium constant for the reaction.

This equation may be considerably simplified by working at the internationally accepted temperature of 298 K, converting to Briggsian logarithms and calculating the appropriate prelogarithmic constant. It is thus reduced to

$$E = E^{o\prime} - 0.05916 \log \frac{[Fe^{2+}]}{[Fe^{3+}]} \quad \ldots\ldots \text{(ii)}$$

for the reaction

$$Fe^{3+} \text{ (complex)} + e^- \rightleftharpoons Fe^{2+} \text{ (aq.)}.$$

The symbol [Fe^{3+}] has been chosen for brevity to represent the concentration of the iron (III) complex.

As is usual with these titrations, a plot of potential, E, against volume of titrant added or fraction of material titrated yields a sigmoidal curve (Figure 1), with the end-point occurring at the inflexion point. If the latter point is measured accurately, and the standard reduction potential of the titrant is known, that for the unknown compound may be determined (1). Reference to the paper cited shows that the theory is a little complex and, in this instance, a simpler approach may be adopted because the concentration of iron in both forms may be calculated at any point during the titration. A plot of measured potential against the logarithm of the ratio of their concentrations yields a straight line, the analysis of which is much simpler (Figure 2). Both intercepts may be used to calculate a value for E° as follows: a) along the potential axis the intercept occurs at the point E = E°'; b) along the abscissa the intercept occurs at E = 0 and E°' is numerically equal to the logarithmic function at this point (Figure 2). If a value for the standard potential of the reference electrode is known, a value for the standard potential of the analyte may be calculated by appropriate addition.

Experimental

1. *Chemicals and Apparatus*

All chemicals used were of analytical grade or better and mixtures of anhydrous benzene and ethanol were used as solvents.

Excepting the sodium hydroxide standardization, all titrations were monitored

FIGURE 1

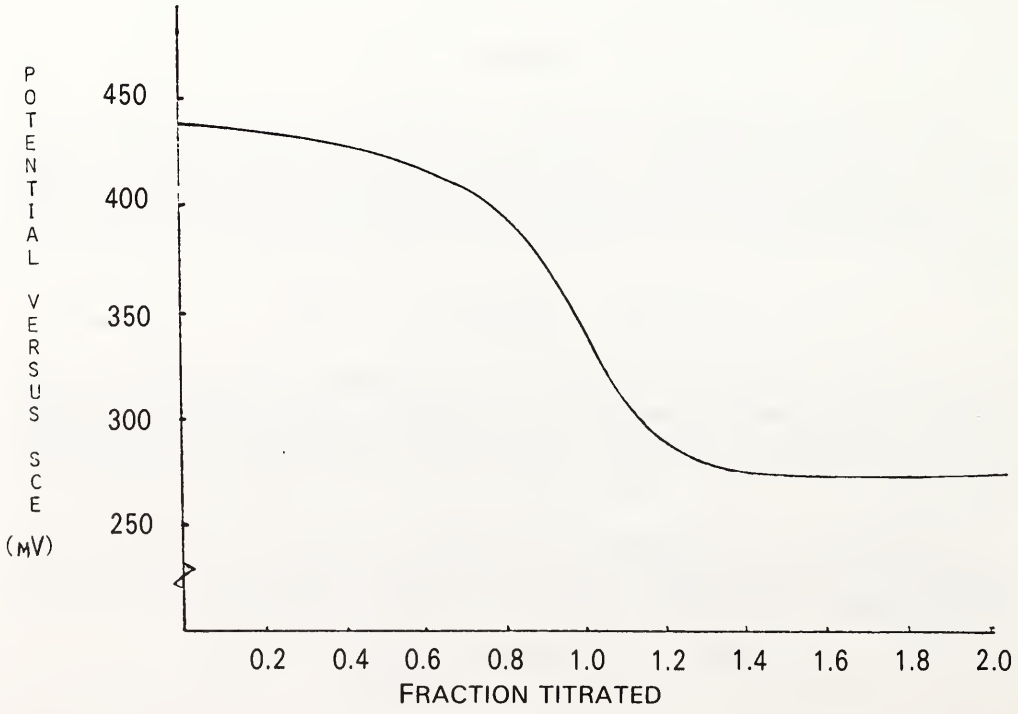
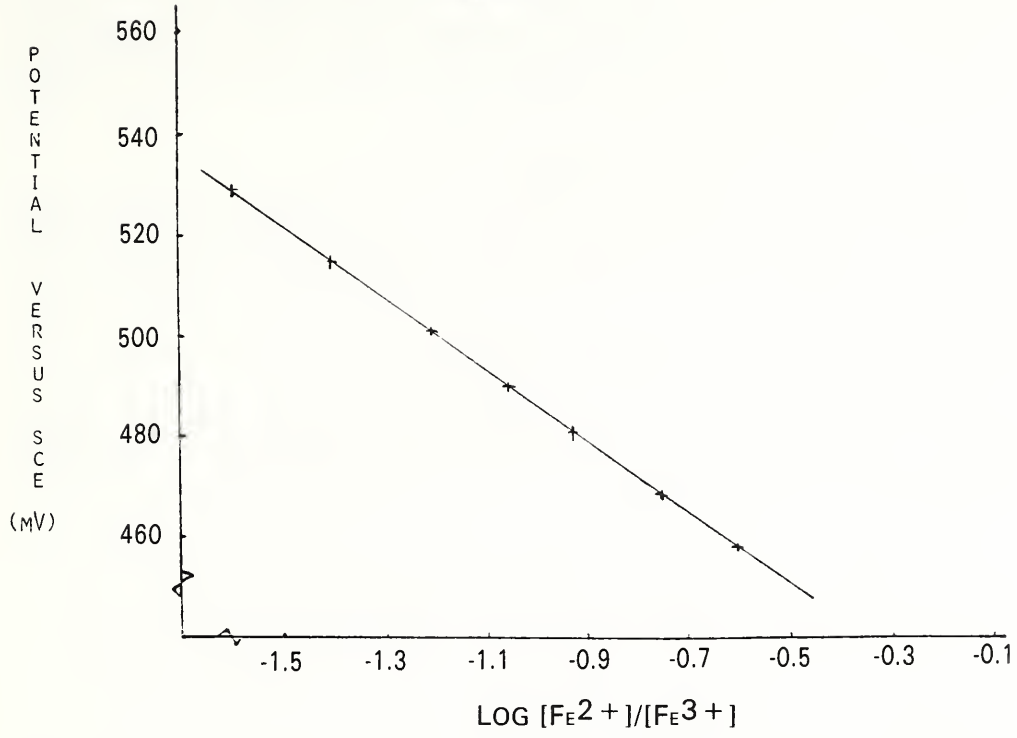


FIGURE 2



potentiometrically. The reference was a saturated calomel electrode standardized against a standard hydrogen electrode, and the responsive electrode was a platinum disc. Experiments were conducted in a thermostatted cell at 25.0°C ± 0.1°C and potentials were measured with a Fisher Accumet Model 420 Digital pH/Ion Meter. All glassware was appropriately calibrated by weighing.

2. *Synthesis of Tetramethylammonium Tetrachloroferrate (III) (4)*

Anhydrous ferric chloride (16.25 g., 0.1 mol.) was dissolved in an anhydrous mixture of benzene (100ml) and ethanol (75ml). The solution thus obtained was mixed with one of tetramethylammonium chloride (10.95 g., 0.1 mol.) dissolved in the minimum amount of anhydrous ethanol. The product, an orange-yellow solid, precipitated and was filtered at the pump. It was recrystallized three times from hot ethanol, dried at 110°C and stored in a desiccator over calcium sulfate. A second crop of material could be obtained by evaporation of the mother liquor.

3. *Analysis of the Complex for Iron*

Enough carefully dried potassium dichromate (7) was weighed accurately to make a solution of approximately 0.025 M. Solutions of accurately weighed quantities of the complex were reduced by passage through a Jones reductor, and then titrated with the standard potassium dichromate to either a potentiometric end-point or one of sodium diphenylaminesulphonate (5). Results are contained in Table 1.

4. *Analysis of the Complex for Chloride*

The conventional gravimetric method for chloride with silver ions was rejected in this case because of coprecipitation of ferric hydroxide.

Aliquots of solutions containing accurately known quantities of the complex

were passed through a bed (1 cm^2 x 30 cm) of Rexyn-100 cation exchange resin in the acid form. The hydrochloric acid produced was titrated with carbonate-free sodium hydroxide solution prepared and standardized in the usual fashion (6). Blank titrations on suitable portions of column effluent were also performed. Results are contained in Table 1.

5. *Determination of Standard Reduction Potential*

Stabilized ascorbic acid solution was standardized iodometrically (2) and was then used to titrate suitable quantities of the complex potentiometrically in acid solution. The results are displayed in Figures 1 and 2, and the data shown in Table 2.

Results and Discussions

Table 1 shows the results of analysis and, within experimental error for the method chosen, the results agree with those obtained theoretically for a substance of composition $(CH_3)_4NFeCl_4$.

TABLE 1: *Results of Iron and Chloride Analysis*

Species Determined	Result	Standard Deviation
Iron (20.55%)	20.58%	± 0.03%
Chloride (52.17%)	51.8%	± 0.4%

Reference to Figure 1, which shows a typical result, indicates that plots of potential against volume of titrant added yield sigmoidal curves with well-defined breaks. End-points for titrations could be ascertained either by application of the differential method or that of Gran (3). With regard to the determination of standard reduction potential, Figure 2 shows typical results of a titration, the potential with respect to a saturated calomel electrode being plotted against log $[Fe^{2+}]/[Fe^{3+}]$. All lines produced in this fasion were analyzed statistically and correlation coefficients of the order of 0.999 or better were obtained. Calculation results for typical lines are given in Table 2.

TABLE 2: *Standard Potential Determinations*

Nernst Slope (mV)	E-Intercept (mV)	$E_{1/2}$ep (mV)
-57.5	718	717
-60.2	685	686
-60.8	718	718

Reference to Table 2 shows that the iron (II)-iron (III) couple considered is, to all intents and purposes, perfectly reversible because the Nernst slope is almost exactly that predicted by theory, *ie*: -59.6mV experimentally against -59.1mV theoretically.

Finally, reference to the straight line graphs produced, and statistical analysis of the data yield for the reaction

$$Fe^{3+} \text{ (complex)} + e^- \rightleftharpoons Fe^{2+} \text{ (aq.)},$$

$$E^\circ = +0.707V \pm 0.017V.$$

Acknowledgment

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ABSTRACTS

Size Distributions of Dune Pond Plankton: The Role of Herbivory. Ann M. Bergquist and Stephen R. Carpenter, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.—We tested contrasting explanations for size shifts in limnetic phytoplankton and zooplankton. If the size structure of a phytoplankton assemblage influences the zooplankton size distribution, then as phytoplankton increase in size from spring to summer, the zooplankton size structure should shift from large to small, since larger zooplankton are more inhibited by large colonial and filamentous algae. Alternatively, if the zooplankton size distribution regulates the phytoplankton size structure, the opposite trend is expected. A shift to smaller zooplankton encourages smaller phytoplankton because smaller zooplankters have lower feeding rates and higher nutrient regeneration rates that favor small, rapidly growing algae. A shift to larger zooplankton favors larger phytoplankton that are not readily available to herbivores.

These contrasting hypotheses were tested using seasonal phytoplankton and zooplankton size distributions from two intradunal ponds at the Indiana Dunes National Lakeshore. Large cladocerans dominated the spring zooplankton sample in pond A, but by July the zooplankton assemblage consisted of much smaller herbivores. In pond B the opposite trend was observed. Phytoplankton size increased in pond B: small diatoms, *Scenedesmus* and Chlorococcales were succeeded by *Ceratium*, *Dinobryon*, *Fragillaria* and others. The phytoplankton genera in pond A did not change markedly, but the relative abundances of smaller individual and colonial forms increased. Our results do not support the hypothesis that phytoplankton influence zooplankton size distributions, but are consistent with the hypothesized effect of zooplankton on phytoplankton size distribution.

Scale of Spatial Pattern: Four Methods Compared Using Simulations and Field Data. Jack E. Chaney, Stephen R. Carpenter and Nancy J. McCreary, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.—Four methods of pattern analysis were compared: Nested ANOVA (Ann. Bot. *16*: 293), two-term local variance (J. Ecol. *61*: 225), random pairing (Vegetatio *29*: 135) and spectral analysis (Ann. Rev. Ecol. Syst. *6*: 189). Simulated patterns were: (1) fixed patches separated by gaps equal to patch length; (2) fixed patches with gaps 5X patch length; (3) patches of fixed length, gaps of random length; (4) patches and gaps of random length. The first peak of the variance-block size graph was used instead of the highest peak because it was more accurate in detecting correct patch size. Nest ANOVA and two-term local variance placed the patch size of patterns 2 - 4 between patch and gap size. Spectral analysis maximized variance at the mean full-cycle length. Even in the most variable simulations, random pairing showed a peak at the mean patch size.

Submersed vegetation was sampled using SCUBA along transects parallel to the depth contours. The random pairing method indicated a patch size of 40 cm. Fish nests of pumpkinseed sunfish. (*Lepomis gibbosus*) are approximately the proper size to generate this pattern.

Algal Symbiosis in a Freshwater Flatworm, *Dalyellia viridis.* RICHARD W. GREENE, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.——The green color of the freshwater neorhabdocoel, *Dalyellia viridis,* results from the presence of large numbers of green unicellular algae distributed in its integument. Many other genera of freshwater organisms possess similar green symbionts (e.g. *Paramecium, Stentor, Ophrydium, Vorticella, Spongilla*), but *Dalyellia* differs in that the algae are situated intercellularly. Preliminary evidence suggests that the algae in all of the common associations mentioned may be the same, or at least very closely related. Thus, if the algae are functional, *Dalyellia* presents a unique opportunity to study the influence of the host on a symbiotic alga about which much is already known.

Oxygen exchange in *Dalyellia* has been investigated under light and dark conditions and with the photosynthesis inhibitor DCMU. Results show that oxygen is produced in the light in excess of the consortium's requirements. When DCMU is present, oxygen production ceases.

Studies of carbon-14 uptake show that *Dalyellia* incorporates several times more carbon in the light than in dark controls. Evidence, therefore, points to the algae as being functional endosymbionts. Photosynthetic products of the association are currently being analyzed by radio-chromatography, and nutritional relations of the partners also are being pursued.

Patterns of Allocation in *Podophyllum peltatum* L. (Berberidaceae) DEBORAH A. KEGEL, Indiana University, Bloomington, Indiana 47405.——Mayapples, summer-persistant perennial herbs of the eastern deciduous forests, exhibit both clonal growth and sexual reproduction. Ramet allocation patterns were examined through dry weight studies. Sexual shoots were associated with larger rhizomes. Seasonal changes in the dry weight of the old and new rhizome sections were found to differ between sexual and vegetative shoots. The relationship between fruit production and rhizome growth is examined. The role of the rhizome as a storage organ influencing growth patterns is discussed.

A Technique for Measuring Ammonia Volatilization from Surface Application of Urea to *Dactylis glomerata* L. Pasture. J. W. LIGHTNER, C. L. RHYKERD, D. B. MENGEL, L. E. SOMMERS and G. E. VAN SCOYOC, Department of Agronomy, Purdue University, West Lafayette, Indiana 47907.——A field study was conducted to determine NH_3 losses of urea and urea-based fertilizers when surface applied to orchardgrass (*Dactylis glomerata* L.) pasture located on the Purdue Agronomy Farm, West Lafayette, Indiana. The NH_3 collection system consisted of a vacuum pump, a chemical (0.5 H_2SO_4) trap to capture NH_3, and a volatilization chamber. (Hoff et al. 1981. J. Environ. Qual. 10:90-95). This technique was developed to measure ammonia voltilization in a conventionally tilled corn field.

The volatilization chamber, which provided the microplot, consisted of a PVC cylinder 30-cm I.D. by 25-cm long by 32.4-cm O.D. The cylinder was inserted into the soil leaving approximately 2-4 cm of the cylinder exposed above the soil surface. A plexiglass lid sealed the microplot off to the environment for short intervals during the day when NH_3 loss was actually being measured. Between measurements the lid was removed to allow for normal environmental conditions.

On one side of the volatilization chamber there were five transfer ports con-

nected by tygon tubing to an NH_3 trap, and on the other side of the cylinder there was an inlet port which opened to the ambient environment. The total NH_3 loss was calculated by integrating the rate of loss over time.

A manifold system made of 2-cm CPVC water line with a regulator valve at each of 7 inlets permitted the simultaneous sampling of seven microplots. Air flow rate was in the range of 25-30 exchange volumes per minute per microplot. Volatilization studies on the orchardgrass pasture revealed that this technique has potential for measuring NH_3 losses from surface applied urea and/or ureabased fertilizer to grass pastures.

Competition and Coexistence in Submersed Freshwater Perennials. NANCY J. MCCREARY, STEPHEN R. CARPENTER and JACK E. CHANEY, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.——Competition between two submersed perennials was studied *in situ* in Roach Lake (Gogebic Co., MI), using a de Wit replacement series. *Juncus pelocarpus* forma *submersus* Fassett and *Eleocharis acicularis* (L.) R. & S. were placed in the lake in pots subjected to two light levels, two sediment types, and two densities. The species were combined at five ratios and replicated four times for each treatment. The response variable was final yield on a dry weight basis.

Effects of light, density, and sediment on yield in pure culture were highly significant ($P = 0.0003$). Full data set analysis showed differences in species' yield ($P = 0.01$), and between environmental treatments ($P < 0.0001$). Analyses of species and ratio within each environmental treatment showed no significant differences in the control, sediment, or shading treatments, however, the density treatment showed a significant ratio effect ($P < 0.0001$). Relative Yield Totals approximated one in all treatments (0.84 to 1.0; SD = 0.29 to 0.4). Ratio diagrams for all four treatments indicate stable coexistence throughout the season. While *Eleocharis* grew more rapidly in these experiments, each species is limited more strongly by conspecifics than by neighbors of the other species.

An Analysis of Dispersal in an Unmanipulated Population of *Peromyscus leucopus*. MARK MINER and DAVID T. KROHNE, Department of Biology, Wabash College, Crawfordsville, Indiana 47933.——The demographic characteristics and the consequences of dispersal were studied in a population of *Peromyscus leucopus* with an extensive live-trapping system. Dispersal events up to 1 km (minimum straight line distance) could be detected with this system. Although the absolute number of dispersers increased with density, relative indices showed no correlation with density. Dispersers were a random sample of residents with respect to age structure but the sex ratio was significantly more male-biased than that of residents. Dispersers appeared to suffer high mortality associated with the move and the establishment of residency in the new area. Once residency was established however, there was no difference in mortality rate between residents and newly arrived dispersers. Dispersal was not preferentially directed to regions of lower density. It is suggested that dispersal in this species is more intimately related to social interactions and the avoidance of inbreeding than to demography.

Replacement of *Ulmus americana* **L. in an Old-Growth Central Indiana Woods.** G.R. PARKER and D.J. LEOPOLD, Department of Forestry and Natural Resources, Purdue University, West Lafayette, Indiana 47907.——In 1926 all trees ($\geq$ 10 cm dbh) were permanently tagged and mapped in a 20.6 hectare, old-growth tract of timber in Randolph County, Indiana. The central 8.5 hectares were resurveyed in 1976. The relative basal area and relative density of *Ulmus americana*, American elm, at this time was 4.2% and 12.9%, respectively. By 1976, 94.5% (172 individuals) of the tagged

U. americana were dead. Species which grew into the gaps created by dead American elm were (out of 451 ingrowth trees ≥ 10 cm dbh) *Ulmus americana* (23%), *Acer saccharum* (14%), *Celtis occidentalis* (9%), *Carya ovata* (8%), *Fraxinus americana* (6%), *Fagus grandifolia* (4%), and other species (36%). Though Dutch elm disease and phloem necrosis has killed most American elms that existed in the initial survey, the density had increased three-fold by 1976. Precocious reproductive maturity and shade tolerance has allowed *U. americana* to exist in this old-growth forest, its presence generally restricted to smaller diameter individuals.

An Analysis of the Community Gradient Between an Aspen-Maple Upland Forest and an Alder-Cedar Swamp Forest in Northern Lower Michigan. CHRIS PETERSON and EDWIN R. SQUIERS, Departments of Biology and Environmental Science, Taylor University, Upland, Indiana 46989.——The structure of the plant community in the ecotone between an aspen-maple upland forest and an alder- cedar swamp forest was studied at the Grass River Natural Area, Antrim County, Michigan. Basal area, density, and frequency data were collected by taxonomic species from circular quadrats of 100 square meters each, spaced at 30 meter intervals along six parallel transects between the two community types. Elevation and depth to ground water was also recorded at each quadrat. The ecological distance between quadrats was assessed using a variety of indices of diversity and community similarity, Bray-Curtis ordination, and principle component analysis. The results indicate that the changes along the ecocline are complex and species specific with dominant species such as large-toothed aspen, northern white cedar, and braken fern displaying relatively sharp distributional boundaries while the abundance of many of subdominants changes very gradually.

An Eco-Inventory of the Grass River Natural Area, Antrim County, Michigan. EDWIN R. SQUIERS, MARTHA SITLER, MARK BREEDERLAND and CHRIS PETERSON, Departments of Biology and Environmental Science, Taylor University, Upland Indiana 46989.——This presentation reviews the findings of a three-year ecological inventory of the Grass River Natural Area. The 1000 acre preserve surrounds the Grass River, a part of the watershed that drains east central Antrim County, Michigan. The river flows south then west across an outwash plain of Wisconsin glacial till between Lake Bellaire and Clam Lake. Several cold water trout streams enter the river as it meanders through sedge meadows, northern white cedar swamps and a variety of other northern plant associations. The area displays a flora of more than 400 species highlighted by the presence of sundew, pitcher plants, and a variety of orchids, ferns and mosses. Bald eagle, osprey, mink, and otter fish the river while bear, bobcat and coyote roam the surrounding swamp forest.

Summary data will be presented from each of the six major components of the study: 1) Ecosystem Descriptions, an evaluation of the terrestrial, wetland and aquatic habitats; 2) Ecosystem Mapping, vegetation, hydrology, soils, etc.; 3) Floristic Inventory, vascular and nonvascular species; 4) Faunistic Inventory; 5) Water Quality, a chemical and biological evaluation of the major aquatic systems; and 6) Identification and Documentation of Ecologically Sensitive or Otherwise Noteworthy Areas. The segments of the study dealing with trophic dymamics in the sedge meadow and the aquatic ecosystems will be reviewed in detail.

Use of *Aeromonas hydrophila* as an Indicator of Trophic State in Indiana Lakes. CARL E. WARNES, Department of Biology, Ball State University, Muncie, Indiana 47306.——Lakes and reservoirs throughout Indiana were sampled for densities of *A. hydrophila* to determine if their numbers could be used to indicate lake

trophic status. Reservoirs sampled included Bass and Holiday (Porter Co), Brookville, Eagle Creek, Lemon, Morse, Prairie Creek, and Salamonie. Cedar (Lake Co), Crooked (Whitely Co), Maxinkuckie, Syracuse, Waubee, Wawasee, and Lake of the Woods (Marshall Co) were the natural lakes sampled. Comparisons of trophy were made on the basis of Carlson's Trophic State Indices (TSI) and that of the Indiana State Board of Health (ISBH). Correlation with *A. hydrophila* numbers to these calculated values were used to indicate the validity of the hypothesis. Correlations to various physical-chemical parameters, season and lake sites were also performed.

Rats and Mice Transmit More than 200 Zoonotic Pathogenic Organisms. WALTER WEBER, Indianapolis, Indiana.——A review of medical literature (human and veterinary) has indicated more than 200 documented zoonotic pathogenic organisms affecting man and domesticated animals have been identified with rats and mice. These include 105 bacterial, 8 rickettsial, 10 mycotic, 22 viral, 14 protozoal, 14 cestodes, 18 nematodes, and 29 trematodes. Although hundreds of arthropods have been found on rodents, forty two ectoparasites have been identified with the zoonotic organisms. These include 8 species of mites, 19 ticks, 11 fleas, and 5 other insects. Part of the life cycle of these organisms may be spent in the blood stream, internal organs, saliva or excretions. The disease organisms may be spread by bites, ectoparasites, urine, feces, oral, ocular, or nasal secretions. Since the pathogens are often in the blood and vital organs of the rodent, an animal eating them may aquire the disease. The significance of rats and mice in transmission of pathogenic organisms is considerable and complex. Many of these facts have been and others are being investigated by scientists throughout the world.

Limnological Characteristics of a Southern Indianan Lake During Manual Removal of Macrophytes Followed by Addition of a Photosynthetic Inhibiting Dye. RICHARD L. WHITMAN, Department of Biology, Indiana University-Northwest, Gary, Indiana 46408.——During summer 1979 a limnological investigation was conducted on Reflection Lake located on the campus of Indiana State University-Evansville. Intensive manual removal of submerged aquatic macrophytes occurred between June 10 and July 16. Twelve liters of Aqua Shade® (a photosynthetic inhibiting dye) was added to the lake between July 17 and July 23 and an equivalent amount on Aug. 1. Both manual removal and Aqua Shade addition were successful in removing unwanted vegetation, but the former technique may require excessive use of manpower.

Phosphates, nitrates, nitrites, chlorides, silicates, pH, hardness, dissolved oxygen, temperature, chlorophylls, phytoplankton and fish population were analyzed during lake treatment. Following the cessation of macrophyte removal and initiation of dye treatment: epilimnetic oxygen levels rose, as did filamentous blue greens, chlorophyll b, pH, and littoral nitrites. Temporary increases in phosphates, nitrates, and turbidity occurred with dye application, while total hardness generally decreased. Fish population analyses did not indicate an adverse effect on bluegill and largemouth bass from the above described treatments.

The Use of Sediment Oxygen Demand in Stream Sediment and Water Quality Assessment of Salt Creek, Porter County, Indiana. RICHARD L. WHITMAN and EUGENE J. FARON. Department of Biology, Indiana University-Northwest, Gary, Indiana. 46408——Oxygen uptake of stream sediments and its relationship to bottom fauna, developing fish eggs, and chemical interactions has received increasing attention. In this study we measured oxygen uptake rates of Salt Creek

sediments using Gilson Respirometry during July, Aug., and Sept. 1980 and April 1981. Two locations were selected upstream of the Sewage Treatment Plant and six below it. The technique described is relatively simple and uses conventional equipment and methodology. Replication is somewhat higher than reported for other more traditional oxygen demand indices (BOD, COD, etc.), although station transects showed considerable variation due to heterogeneous sediment composition. Sediment organic content, stream water temperature and nitrates were positively correlated with oxygen uptake rates, while surface water velocity, turbidity, total suspended solids, pH, and alkalinity correlated in a negative manner ($p = 0.05$). Oxygen uptake is significantly higher in sediments just below the treatment plant ($p = 0.05$). Sediment oxygen uptake rates in conjunction with 48 hours diurnal oxygen monitoring give a reasonably good description of dissolved oxygen relationships in a stream subjected to perturbations from organic loading.

An Annotated List of the Fishes of Eagle, Stotts and Rattlesnake Creeks in Central Indiana

WILLIAM L. FISHER
Water Resources Laboratory, University of Louisville,
Louisville, Kentucky 40292
and
JAMES R. GAMMON
Department of Zoology, DePauw University,
Greencastle, Indiana 46135

Introduction

Since the extensive statewide ichthyofaunal survey of Gerking (3), relatively few studies have focused on the fishes of central Indiana streams. Distributional studies such as these are important in evaluating the impact of man's activities on the environment as well as assessing long term changes in fish distribution.

A study of the fishes of Eagle Creek, Stotts Creek and Rattlesnake Creek, all tributaries of the west fork of the White River, was conducted during the summers of 1978, 1979 and 1980 as part of the Model Implementation Program (MIP). The project, initiated by the U. S. Environmental Protection Agency and the Department of Agriculture, utilized the resources of various state agencies and universities to monitor and model the response of stream biota and water quality to institution of best agricultural land management practices within the Eagle Creek and Stotts Creek watersheds. Rattlesnake Creek was chosen to serve as a control stream for the project.

The purpose of this report is to provide new and updated distributional information on the fishes of these three streams, and to compare their faunal composition.

Materials and Methods

Fish were collected twice each summer during 1978, 1979, and 1980 from permanent stationis established on Eagle Creek, Stotts Creek and Rattlesnake Creek, unless otherwise noted (see STUDY AREA). Each station consisted of a pool and its adjacent upstream riffle. Most of the collections were make using a 30-foot electroseine powered by a 1350-watt A.C. generator, with additional samples obtained by using a one-eighth-inch-mesh nylon seine, and a D.C.-powered electrofishing apparatus mounted on a john boat. Most of the fish were identified, weighed and measured in the field to prevent the reduction or elimination of less common species. Representative voucher specimens of each species were preserved and are deposited in the DePauw University museum. Laboratory idientifications of the preserved specimens were make using the keys of Trautman (8), Pflieger (5) and Smith (7). One hundred nine individual collections were made during the three-year study.

Study Area

Eagle Creek is located northwest of Indianapolis in central Indiana, and lies within four counties: Hendricks, Boone, Hamilton and Marion (Fig. 1). The study area included that portion of the watershed which drains into Eagle Creek reservoir; encompassing about 419 km^2 (103,000 acres). Land usage within the water-

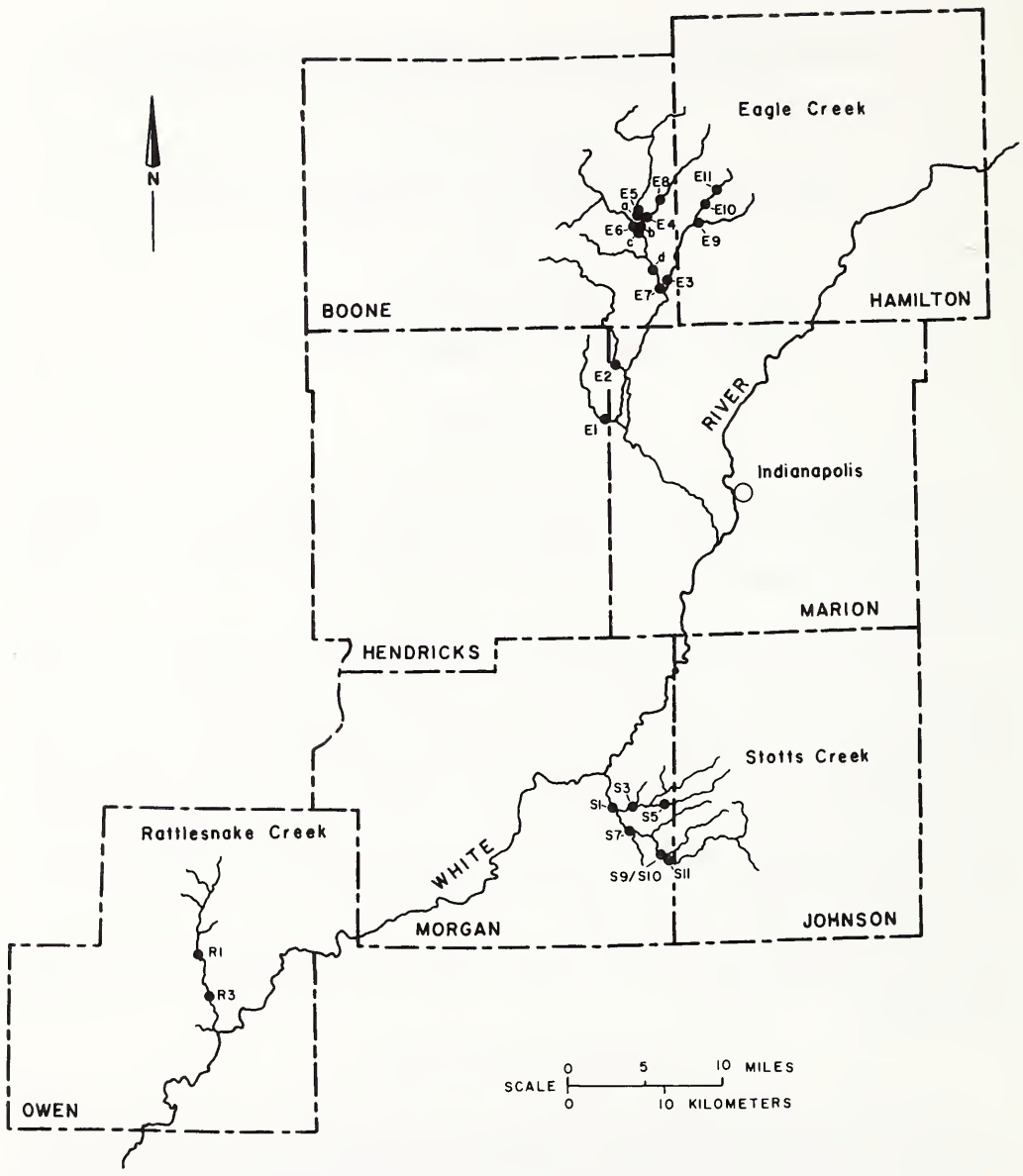

FIGURE 1. *Map of the study streams showing the location of the sampling stations.*

shed is dominated by croplands which comprise 70% of the area. Livestock is an important farming practice and numerous feedlots are found within the watershed. About 5% of the land is presently developed as urban and transportation areas, and forests cover 13.% of the watershed (Indiana Heartland Model Implementation Project, 1980 status report).

Seven permanent sampling stations were established on Eagle Creek and its tributaries. Eight additional stations were sampled only once during the study. The stations were similar in physical dimensions and are generally characterized by having relatively stable substrates, good riparian habitat and constant flow. Below are brief descriptions of the Eagle Creek collecting stations including: station number, stream order, section, township and range designations, stream location descriptions, and various physical and habitat characteristics:

E1. School Branch; order II; sec. 17, T.16N., R.2E.; morphometry—$\bar{X}$ length 45 m (42-49), $\bar{X}$ width 5.7 m (2.7-7.0), $\bar{X}$ depth 24 cm (3-100); substrate—sand-granule-

silt; riparian—forest with trees and shrubs lining moderate to steeply sloping banks, dense canopy.

E1. Fishback Creek; order III: sec. 29, T.17N., R.2E.; morphometry—$\bar{X}$ length 42 m (33-53), $\bar{X}$ width 5.7 m (2.7-9.0), $\bar{X}$ depth 26 cm (4-90); substrate—cobble-pebble-sand; riparian—forest with trees and grasses lining gradually sloping banks, moderate canopy; comments—pool and riffle 0.1 miles upstream from bridged sampled in 1978.

E3. Little Eagle Creek; order III; sec. 25, T.18N., R.2E.; morphometry—$\bar{X}$ length 54 m (47-68), $\bar{X}$ width 10.0 m (6.0-14.7), $\bar{X}$ depth 25 cm (2-92); substrate—pebble-cobble-sand; riparian—forest with trees and shrubs lining moderate to steeply sloping banks, dense canopy.

E4. Finley Creek; order II; sec. 11, T.18N., R.2E.; morphometry—$\bar{X}$ length 27 m (26-31), $\bar{X}$ width 4.3 m (2.0-5.7), $\bar{X}$ depth 26 cm (3-60); substrate—silt-cobble-pebble, sewage fungus occasionally blanketed the riffles; riparian—trees lined gradually sloping banks, moderate canopy; comments—active landfill operation immediately upstream from station.

E5. Eagle Creek; order III; sec. 3, T.18N., R.2E.; morphometry—$\bar{X}$ length 36 m (30-39), $\bar{X}$ width 10.7 m (7.3-13.7), $\bar{X}$ depth 40 cm (4-110); substrate—cobble-pebble-sand, rooted aquatic plants in riffles; riparian—cropland with trees and brush lining gradual to moderately sloping bank, moderate canopy.

E5a-d. Four 0.17 km reaches of the mainstem of Eagle Creek were sampled on 15 September 1978, using a D.C. powered electroshocker mounted to a boat. The stations included: E5a above the mouth of Finley Creek, E5b below the mouth of Finley Creek, E5c below the mouth of Mounts Run, and E5d near O'Neal Avenue bridge (Sec. 16, T.18N., R.2E.). Habitat data were not recorded.

E6. Mounts Run; order III: sec. 10, T.18N., R.2E.; morphometry—$\bar{X}$ length 27 m (26-28), $\bar{X}$ width 17.7 m (1.0-8.3), $\bar{X}$ depth 31 cm (5-85); substrate—pebble-granule-sand; riparian—willow shrubs and a few trees lined gradual to steeply sloping banks, sparse canopy.

E7. Eagle Creek; order IV; sec. 35, T.18N., R.2E.; comments—seined once during a low flow period in the summer of 1980. No habitat data were recorded.

E8. Finley Creek; order I; sec. 35, T.19N., R.2E.; morphometry—$\bar{X}$ length 38 m (33-37), $\bar{X}$ width 15.3 m (3.7-5.7), $\bar{X}$ depth 20 cm (8-50); substrate—sand-cobble-silt; riparian—corpland with shrubs and some trees lining moderately sloping banks, sparse canopy.

E9. Drainage ditch above county road bridge, 1.7 miles SW of Eagleton, order II; sec. 8, T.18N., R.3E.; morphometry—length 24 m, $\bar{X}$ width 2.3 m (1.3-3.7), $\bar{X}$ depth 14 cm (5-30); substrate—silt-mud-sand; riparian—cropland with willow shrubs and grass on steeply sloping banks forming dense canopy; comments—4 seine hauls were made on 4 August 1978.

E10. Little Eagle Creek; order II; sec. 33, T.19N., R.3E.; comments—collections were made on 9 July 1980. No habitat data were recorded.

E11. Little Eagle Creek; order II; sec. 5, T.18N., R.3E.; comments—collections were made on 23 July 1980. No habitat data were recorded.

The Stotts Creek watershed near Martinsville, Indiana, lies within Morgan and Johnson counties and drains about 212 square kilometers (52,461 acres). Croplands account for about 58% of the land use in the watershed whereas forests comprise nearly one-third of the total area.

Five permanent sampling stations on Stotts Creek and its tributaries were sampled twice each summer during the study with six additional sites being sampled only in 1978. With the exception of one station (S5), the stream bottom substrate was dominated by sand, granule and pebble particle sizes (0.06-64.0 mm diameter). The combination of small size substrate particles and frequent flooding resulted in major changes in the stream topography in which some of our previously sampled pools were reduced to shallow raceways and riffles.

A description of the sampling stations is given below:

S1. Stotts Creek; order V; sec. 21, T.12N., R.2E.; morphometry—$\overline{X}$ length 48 m (43-52), $\overline{X}$ width 7.3 (3.3-9.3), $\overline{X}$ depth 26 cm (13-104); substrate—granule-sand-pebble; riparian—trees and brush on gradual to moderately sloping bank, moderate canopy; comments—adjacent upstream pool and riffle (S2) were sampled twice in 1978.

S3. North Prong Stotts Creek; order IV; sec. 22, T.12N., R.2E.; morphometry—$\overline{X}$ length 42 m (39-47), $\overline{X}$ width 6.3 m (4.0-8.0), $\overline{X}$ depth 20 cm (1-77); substrate granule-pebble-sand; riparian—trees, willow shrubs and grasses bordered gradual to steeply sloping banks, moderate canopy; comments—three adjacent pools and riffles were sampled during the study including S4.

S5. North Prong Stotts Creek; order III; sec. 24, T.12N., R.2E.; morphometry—$\overline{X}$ length 48 m (45-50), $\overline{X}$ width 7.0 m (3.3-10.0), $\overline{X}$ depth 26 cm (3-67); substrate—cobble-pebble-silt with soft erodable clay; riparian—trees and willow shrubs on gradual to steeply sloping banks, moderate canopy; comments—adjacent upstream station (S6) was sampled during 1978.

S7. South Prong Stotts Creek; order IV; sec. 27, T.12N., R.2E.; morphometry—$\overline{X}$ length 49 m (42-58); $\overline{X}$ width 6.3 m (4.0-11.7), $\overline{X}$ depth 24 cm (3-83); substrate—pebble-granule-sand; riparian—trees and grasses on steep to gradual sloping banks, dense canopy; comments—massive fish kill noted 26 July 1978 evidenced by dead bullheads on shore and paucity of fish in collections; adjacent upstream pool (S8) sampled during 1978.

S9/S10. South Prong Stotts Creek; order IV; sec. 1, T.11N., R.2E.; comments—three adjacent pools and riffles were sampled during 1978. The stations had an average length of 45 m, average width of 5.0 m and an average depth of 19 cm. The substrate was dominated by pebble-granule-sand, and the riparian habitat consisted of forest and cropland with trees lining the banks forming a dense canopy. Instream cover, comprised of submerged roots and trees, was abundant.

S11. South Prong Stotts Creek; order III; sec. 1, T.11N., R.2E.; morphometry—$\overline{X}$ length 30 m (25-35), $\overline{X}$ width 5.0 m (3.7-5.7), $\overline{X}$ depth 20 cm (4-49); substrate—sand-granule-pebble; riparian—moderately sloping banks devoid of vegetation due to recent removal by bulldozer, no canopy; comments—not sampled on 1978.

Rattlesnake Creek lies within Owen County and enters the White River near Spencer, Indiana. The drainage area is 65 square kilometers (16,121 acres) with the upper portion of the watershed draining portions of the Owen-Pubnam State Forest, and the lower portion coursing through bottomland fields and influenced by agriculture. Two permanent sampling stations were established on Rattlesnake Creek, with one additional station being sampled only in 1978. The stations are described below:

R1. Rattlesnake Creek; order III; sec. 2, T.10N., R.4W.; morphometry—$\overline{X}$ length 44 m (34-50), $\overline{X}$ width 6.7 m (4.7-7.7), $\overline{X}$ depth 21 cm (3-85); substrate—pebble-granule-sand; riparian—trees and shrubs on steep to moderately sloping

banks, dense canopy; comments—large upstream pool and riffle (R2) were sampled in 1978.

R3. Rattlesnake Creek; order III; sec. 13, T.10N., R.4W.; morphometry—$\bar{X}$ length 65 m (64-66), $\bar{X}$ width 7.3 m (5.3-9.3), $\bar{X}$ depth 32 cm (3-83); substrate—sand-granule-pebble; riparian—pastureland with grasses and mud on steep sloping banks, no canopy; comments—sewage drainpipe draining into upper end of pool.

Annotated List of Fishes

A total of 51 species representing 31 genera and 12 families were collected from the three streams combined during the study. In the list that follows, Eagle Creek is designated as "EC", Stotts Creek as "SC" and Rattlesnake Creek as "RC". Common and scientific names follow Robbins et al. (6).

Petromyzontiade

Ichthyomyzon castaneus Girard. Chestnut lamprey.

EC—no collections. SC—no collections. RC—four adult specimens (235-282 mm TL) were taken on 5 August 1980 from R3.

Twelve ammocete larvae (73-138 mm TL) with continuous dorsal fins and myomere counts ranging from 51 to 55 were taken in 1978 and 1979 from R1, R2, and R3. These specimens were not assigned to a species due to the lack of sufficient keys for larvae of this type.

Lampetra aepyptera (Abbott). Least brook lamprey

EC—no collections. SC—no collections. RC—a total of five ammocetes (112-149 mm TL) were collected at R1, R2, and R3 in 1978, 1979, and 1980. Identification of the specimens was based on myomere counts (57-59) and pigmentation patterns discussed by Vladykiv (9) and Distler (1).

Lampetra appendix (DeKay). American brook lamprey.

EC—no collections. SC—no collections. RC—a total of four ammocete larvae (77-177 mm TL) were taken from all three stations during the 3-year study period. Their identification was based on myomere counts (67-69) and pigmentation patterns also discussed in Vladykov (9) and Distler (1).

Clupeidae

Dorsoma cepedianum (Lesueur). Gizzard shad.

EC—occasional specimens taken from the slow-flowing, deep pools on the mainstem of Eagle Creek (E5, E5a, E5d, and E7) and at E1 which drains directly into the reservoir. Also captured at upstream tributary station, E8, which often runs turbid. SC—three specimens taken from S5, a large upstream pool that is usually turbid. RC—no collections.

Esocidae

Esox americanus vermiculatus Lesueur. Grass pickerel.

EC—common in slow-flowing, weedy pools (E5 and E6), occasional elsewhere and absent from E1, E2, and E3 collections. SC—no collections. RC—commonly taken from R1, 2 specimens collected at downstream station (R3).

Cyprinidae

Campostoma anomalum (Rafinesque). Central stoneroller.

EC—most abundant species collected, taken from most upstream tributary stations, less abundant in mainstem, early summer collections dominated by juveniles. SC—one of the two most abundant species collected, taken from all stations. RC—most abundant minnow species at downstream station (R3), common in upstream (R1) collections.

Carassius auratus (Linnaeus). Goldfish.

EC—no collections. SC—one small specimen and a goldfish-carp hybrid collected from S9/S10. A large goldfish hatchery is located on Clear Creek, just downstream from Stotts Creek. RC—no collections.

Cyprinus carpio Linnaeus. Common carp.

EC—taken mainly from the large pools of Eagle Creek (E5, E5a-d), also captured at E1, a direct tributary of reservoir, and E6, the first deep pool upstream from mainstem Eagle Creek. SC—one 1500 g specimen taken from upstream station (E5) following a flood. RC—no collections.

Ericymba buccata Cope. Silverjaw minnow.

EC—occasional specimens collected from all upstream tributary stations, none taken from mainstem Eagle Creek. SC—fourth most abundant minnow species collected, taken from all stations and typically found in association with bluntnose minnows and common stonerollers. RC—taken in greater abundance from downstream station (R3) than upstream stations (R1, R2).

Hybopsis amblops (Rafinesque). Bigeye chub.

EC—no collections. SC—no collections. RC—commonly collected at upstream stations (R1, R2), occasional at R3.

Notropis chrysocephalus (Rafinesque). Striped shiner.

EC—common in tributary stations with clear water, moderate current, and cobble-pebble substrate (E2, E3, E5, and E6), occasional elsewhere. SC—fifth most abundant minnow species, taken from all stations. RC—second most abundant minnow species collected, found equally abundant at upstream and downstream stations.

Notropis spilopterus (Cope). Spotfin shiner.

EC—one specimen collected in 1979 from E2. SC—occasionally captured at all stations except S11, most abundant at S1 and S10. RC—occasional specimens, more abundant at R3.

Notropis stramineus (Cope). Sand shiner.

EC—no collections. SC—common in collections from S1, S3, and S4, occasional elsewhere. RC—common at downstream station (R3), occasional upstream.

Notropis umbratilis (Girard). Redfin shiner.

EC—common only in collections from E3 where breeding schools were observed over sunfish spawning nests, occasional elsewhere. SC—occasional specimens

from all stations. RC—commonly taken from upstream stations, 1 specimen collected at R3.

Notropis volucellus (Cope). Mimic shiner.

EC—no specimens. SC—occasional specimens collected from downstream stations S1 and S3. RC—three specimens were collected, one from R1 and two from R3.

Phenacobius mirabilis (Girard). Suckermouth minnow.

EC—three specimens collected, two from E2, the other from E6. SC—occasional specimens taken from S1 and S3. RC—taken mainly from R1, a few specimens collected at R3.

Phoxinus erythrogaster (Rafinesque). Southern redbelly dace.

EC—five specimens collected at E6. SC—occasional specimens taken from stations on South Prong, most common at S7 where a subteranean spring enters the pool. RC—no specimens.

Pimephales notatus (Rafinesque). Bluntnose minnow.

EC—third most abundant species collected, taken from all tributary stations and in mainstem. SC—the most abundant species collected, predominantly found in shallow pools at stations S1, S4, S8, and S9/S10. RC—third most abundant minnow species collected, equally abundant at upstream and downstream stations.

Pimephales promelas Rafinesque. Fathead minnow.

EC—one specimen collected from E4. SC—three specimens collected, two from S11, one from S7. RC—no collections.

Rhinichthys atratulus (hermann). Blacknose dace.

EC—no collections. SC—taken from all stations but most common in upstream collections on South Prong. RC—four specimens collected at R1.

Semotilus atromaculatus (Mitchill). Creek chub.

EC—second most abundant minnow species collected, particularly abundant in collections from E1, E4, and E8. SC—third most abundant minnow species collected, taken from all stations. RC—fourth most abundant minnow species collected, generally more abundant upstream at R1.

Catostomidae

Catostomus commersoni (Lacepède). White sucker.

EC—most abundant sucker species taken, 1979 collections dominated by juveniles, especially at E4 (below landfill) where juvenile creek chub and common stonerollers were also very abundant. SC—most abundant sucker species collected, especially at stations S3, S5, S9/S10, and S11. RC—occasionally collected at all three stations.

Erimyzon oblongus (Mitchill). Creek chubsucker.

EC—occasionally taken from all stations except E1, E3, and the downstream mainstem stations. SC—one specimen taken from S5 in 1979. RC—three specimens collected from R1.

Hypentelium nigricans (Lesueur). Northern hog sucker.

EC—no collections. SC—second most abundant sucker species collected, common in riffles and raceways of downstream stations S1, S2, S3, and S4. RC—the most abundant sucker species collected, taken mainly from the riffle-raceway habitat of R3.

Minytrema melanops (Rafinesque). Spotted sucker.

EC—occasionally taken from large pools of mainstem stations, one specimen taken from upstream station E8. SC—no collections. RC—no collections.

Moxostoma duquesnei (Lesueur). Black redhorse.

EC—no collections. SC—commonly taken in association with golden redhorse from the raceways and deep pools of Stations S1, S3, S8, and S9/S10, absent from collections at S11. RC—common in collections from R3 and in deep pool of R2, a few specimens taken from R1.

Moxostoma erythrurum (Rafinesque). Golden redhorse.

EC—no collections. SC—more abundant in our collections than black redhorse, with which it was always found to occur. RC—occasionally taken with black redhorse which were two times more abundant in our collections.

Ictaluridae

Ictalurus melas (Rafinesque). Black bullhead.

EC—occasionally captured at all stations except E2 and E6, most abundant at E1 which is just upstream from the backwater of Eagle Creek reservoir. SC—three specimens collected, two from S5 and one from S11. RC—no collections.

Ictalurus natalis (Lesueur). Yellow bullhead.

EC—occasionally collected at all stations except E1 and E5. One-half as abundant as black bullheads in our collections. SC—occasionally taken from all stations except S11. RC—no collections.

Noturus miurus Jordon. Brindled madtom.

EC—no collections. SC—occasionally collected in pools from all stations except S5, S9/S10, and S11. RC—no collections.

Cyprinodontidae

Fundulus notatus (Rafinesque). Blackstripe topminnow.

EC—occasional specimens from upstream stations E10 and E11. SC—no collections. RC—no collections.

Atherinidae

Labidesthes sicculus (Cope). Brook silverside.

EC—a few specimens collected from Stations E1 and E7. SC—no collections. RC—no collections.

Centrarchidae

Ambloplites rupestris (Rafinesque). Rock bass.

EC—common in large pools of mainstem stations and E6. SC—no collections. Gerking (4) reported this species from Stotts Creek in the only other published study of this stream. RC—the second most abundant sunfish taken in our collections, equally abundant at upstream and downstream stations.

Lepomis cyanellus Rafinesque. Green sunfish.

EC—taken from all tributary stations except E9, E10, and E11. Common in collections at stations E2 and E3, but sparce in mainstem collections. SC—the most abundant sunfish collected, taken from all stations and particularly abundant at S5. RC—occasionally collected at upstream and downstream stations.

Lepomis gulosus (Cuvier). Warmouth.

EC—no collections. SC—one specimen taken from station S5. RC—no collections.

Lepomis macrochirus Rafinesque. Bluegill.

EC—commonly taken from tributary stations, less abundant in mainstem collections. SC—commonly collected from all stations, most abundant at S5. RC—occasional collections at upstream and downstream stations.

Lepomis megalotis (Rafinesque). Longear sunfish.

EC—the most abundant sunfish collected, common in mainstem collections and at station E3. SC—occasionally collected at all stations except S11. RC—the most abundant sunfish collected, considerably more numerous at downstream station R3.

Lepomis microlophus Günther). Redear sunfish.

EC—one specimen collected from station E5c on Eagle Creek below Mounts Run. SC—one specimen taken from station S5. RC—two specimens collected, one from station R1 the other from R3.

Micropterus dolomieui Lacepede. Smallmouth bass.

EC—three specimens collected; one 215 mm adult from station E5 and two juveniles from E6. SC—commonly taken from all stations except S7, abundant at S5 where several large adults were collected. RC—occasionally taken from upstream and downstream stations.

Micropterus punctulatus (Rafinesque). Spotted bass.

EC—occasional specimens, mostly juvenile, collected from tributary stations, less abundant in mainstem. SC—juvenile specimens commonly collected from all stations. RC—six specimens taken, one from R3, the rest from upstream stations.

Micropterus salmoides (Lacepede). Largemouth bass.

EC—occasionally taken from all tributary stations, except E9, E10, and E11, absent from mainstem collections except E5 where several large adult specimens were captured. SC—occasional specimens taken from all stations except S9/S10 and S11, most numerous at S5 where several large adults were collected. RC—occasionally collected at both upstream and downstream stations.

Pomoxis annularis Rafinesque. White crappie.

EC—common in mainstem collections, also taken from E1, E6, and E8. SC—four specimens collected at S9/S10. RC—no collections.

Pomoxis nigromaculatus (Lesueur). Black crappie.

EC—no collections. SC—one specimen taken from S9/S10. RC—no collections.

Percidae

Etheostoma blenniodes Rafinesque. Greenside darter.

EC—no collections. Previously reported by Fisher and Gammon (2), however, no voucher specimens were available for verification. SC—common in larger, permanent riffles at S5 and S7, occasional elsewhere. RC—occasionally collected in upstream riffles, a few specimens taken downstream.

Etheostoma caeruleum Storer. Rainbow darter.

EC—common in collections from E5 and E6, occasional or absent elsewhere. SC—abundant in riffles at S3, S5, and S7, occasional elsewhere, absent from S11. RC—the most abundant darter species collected, five times more abundant upstream (R1) than downstream (R3).

Etheostoma flabellare Rafinesque. Fantail darter.

EC—no collections. SC—occasionally collected at all stations except S11. RC—no collections.

Etheostoma nigrum Rafinesque. Johnny darter.

EC—one of the two most abundant darters collected, taken in abundance from sandy pool habitat of E3, E6, and E8. SC—abundant in collections from all stations, especially S7 and S11. RC—abundant; twice as many collected downstream (R3) than upstream (R1, R2).

Etheostoma spectabile (Agassiz). Orangethroat darter.

EC—most abundant darter species collected, taken in abundance from riffles at all stations except E4, E5, and mainstem stations where riffle habitat was not sampled. SC-most abundant of the darters, taken from all stations, and especially numerous at S3, S7, and S11. RC—equally common at upstream and downstream riffles.

Percina caprodes (Rafinesque). Logperch.

EC—commonly taken from E1, E2, and E5 in deep, swift-moving water below riffles, occasionally collected at E6 and E5d. SC—no collections. RC—no collections.

Percina maculata (Girard. Blackside darter.

EC—occasionally collected from E2, E5, and E8 in habitat similar to that of logperch darters. SC—no collections. RC—occasional specimens taken upstream at R1 and R2, one specimen found downstream at R3.

Cottidae

Cottus bairdi Girard. Mottled sculpin.

EC—common in the riffles at stations E1, E6, and E8, occasional or absent

elsewhere. SC—no collections. RC—commonly collected at upstream and downstream sites.

Discussion

Thirty-five species representing 10 families were identified from our collections of Eagle Creek and its tributaries. Five species, the central stoneroller *Campostoma anomalum*, striped shiner *Notropis chrysocephalus*, bluntnose minnow *Pimephales notatus*, creek chub *Semotilus atromaculatus*, and white sucker *Catostomus commersoni*, comprised 71% of the total number of fish caught and 63% of the total weight of the catch.

The fish community of Eagle Creek has changed considerably in the past fifteen years following treatment of the entire drainage with rotenone and the subsequent impoundment of the stream in 1966 (Fisher and Gammon 2). Most evident is the apparent loss of northern hog suckers *Hypentelium nigricans* and golden redhorse *Moxostoma erythrurum* along with several minnow and darter species from the system, and the paucity of smallmouth bass *Micropterus dolomieui*. Factors such as increased agriculturization and urbanization within the watershed have no doubt also played a role in altering the community composition of this stream system.

The collections from Stotts Creek contained 39 species representing six families. Of those thirty-nine, the central stoneroller, striped shiner, bluntnose minnow, creek chub, and white sucker again accounted for three-fourths of the total numbers and two-thirds of the total weight of the catch. Cyprinid species were particularly numerous in Stotts Creek, and in general appeared best able to sustain the large fluctualtions in stream flow and resultant shifting of bottom sediments observed both by us and Gerking (4).

Thirty-five species from seven families were taken in our collections at Rattlesnake Creek. They represented a strikingly different community from that of both Eagle Creek and Stotts Creek. The central stoneroller, striped shiner, bluntnose minnow, and creek chub comprised nearly one-half of the total catch, while two-thirds of the total biomass was attributed to four different species: the northern hog sucker, golden redhorse, black redhorse *Moxostoma duquesnei*, and rock bass *Ambloplites rupestris*. The occurance of three species of lampreys, family petromyzontidae, in our collections further exemplifies the unique character of this stream. Vladykov (9) noted that ammocetes belonging to two or more species could often be found in the same section of a stream.

The compositional similarities and differences of the three streams can be further illustraded in a comparison of the relative abundance of the major families (i.e., cyprinids, catostomids, centrarchids, and all other families combined) (Fig. 2). The biomass of the fish communities of both Eagle Creek and Stotts Creek is dominated by cyprinids; to a lesser extent catostomids, and with relatively few centrarchids. In contrast, Rattlesnake Creek was dominated by catostomids, with cyprinids and centrarchids contributing almost equally to the total biomass of our collections. These compositional differences can be attributed to various factors including: drainage basin size and morphology, geologic and environmental history, land usage, and man's activities within the watershed.

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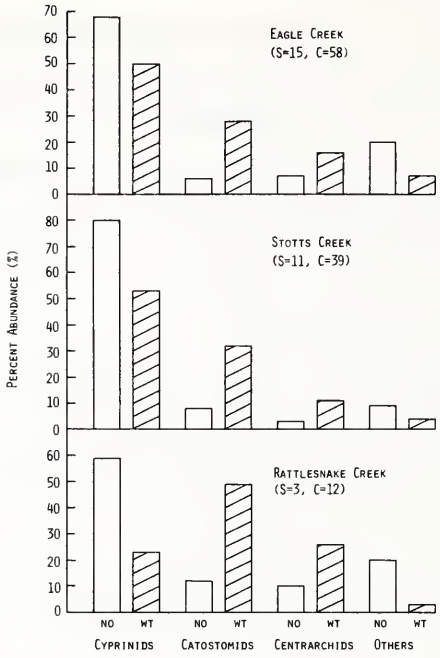

FIGURE 2. *Relative numeric and biomass abundance of the major families of fish of Eagle, Stotts and Rattlesnake Creeks. S denotes number of collecting stations, C denotes number of collections.*

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The Fish Communities of Big Vermilion River and Sugar Creek

J. R. Gammon and J. R. Riggs
Department of Zoology, DePauw University
Greencastle, Indiana 46135

Introduction

The waters of Big Vermilion River and Sugar Creek enter the Wabash River 20 kilometers apart in Vermillion and Parke County, Indiana respectively. In 1979 and 1980 sampling programs of their fish communities were undertaken as part of a long-term study of the middle Wabash River. We wished to (1) determine the nature of the fish communities in the lower portions of each tributary and (2) assess their potential relationship to the Wabash River and its fish community.

The Study Streams

Big Vermilion River and Sugar Creek both flow through wooded valleys on their way to the Wabash River. Big Vermilion River drains 3712 km^2 (1434 mi^2) of largely agricultural land. Its three major tributaries coalesce near Danville, Illinois. Salt Fork and Middle Fork join at River Mile 29 and North Fork enters at RM 23 just upstream from a dam. The Middle Fork is regarded as one of Illinois's finest streams (18) and harbours several unusual species of fish. The upper tributaries of Salt Fork have been the subject of many investigations (9, 10, 11, 15) and suffers domestic sewage from the cities of Rantoul and Champaign-Urbana as well as agricultural runoff. North Fork is somewhat polluted below Lake Vermilion (18).

We investigated the lower 46 km (29 mi.) which, in addition to the waters mentioned, also received drainage from old coal mines and the industrial and domestic wastes from Danville, Ill. (17). Smith (18) stated that this section of the river is "badly polluted . . ., but recovers before it leaves the state." The lower 8 km (5 mi.) exhibits considerable lateral erosion in passing through agricultural fields before joining the Wabash River. The mean rate of fall in the lower reach is about 0.40m/km (2.1 ft/mi).

Sugar Creek is essentially linear in its drainage, flowing about 90 miles from its headwaters in central Indiana westward to the Wabash River and having a mean rate of fall of 0.87m/km (5.1 ft/mi). Its drainage basin is 2100 km^2 (811 mi^2). Diffuse nonpoint agricultural influence occurs in the upper half of the basin and also in the lower 16 km (10 mi), but domestic sewage from Crawfordsville is the only major point-source of influence in the basin. From RM 30 to Rm 10 Sugar Creek flows through a rugged, forested region which includes Pine Hills State Nature Preserve, Shades State Park, and Turkey Run State Park. Some agricultural fields exist in suitable areas, but they are usually well buffered from the Creek by a wooded corridor. The lower 16 km (10 mi.) of Sugar Creek exhibits considerable lateral erosion. A very limited potential for coal mine runoff exists in the extreme lower portion of the stream.

The distribution of fishes in Sugar Creek prior to 1945 is documented by Gerking (7). More recently, Huffaker (8) has examined the fish populations.

Methods

In 1979, eight collecting stations of 0.5 km length were located within the

lower 30 km of the Big Vermilion River. Each of these stations was sampled three times using a Smith-Root Type VI D.C. electrofishing unit which pulsed 120/sec through an electrode system mounted in a 16 ft Jon boat. This same unit was used regularly on the Wabash River, but the low late summer flow of Big Vermilion River made it difficult to move about. Therefore, in 1980 the same apparatus used on Sugar Creek, a battery powered D.C. unit mounted in a rowing canoe was used to collect fish from the same stations and an additional five which extended the study to the lower 46 km.

In 1979, a total of 19 collecting stations were sprinkled through the lower 80 km of Sugar Creek from Carlington to the mouth. This was reduced to 11 stations in the lower 40 km in 1980 from Deer Mill to Cox Ford bridge. Each station was located in swifter sections of the streams with good bottom cover and moderate depths. Each station was sampled three times in both years.

Electrofishing was conducted near shore and cover in a downstream direction. Netted fish were placed in a livewell and measured, weighed, and identified shortly after being collected and then returned to the water. A few small fish of uncertain identification were placed in 10% formalin and returned to the laboratory for later examination. Several taxonomic keys were used to identify fish (20,2,14,19).

The data from each catch was entered into a computer data file and analyzed for the following parameters:

(1) Shannon-Weaver Index of Diversity (H) (12)

$$\bar{H} = - \sum \frac{(ni)}{N} \text{Ln} \frac{(ni)}{N}$$

where n_i = numbers or weight for individual species per km electrofished
N = total number of individuals or total weight
Ln = natural logarithm

(2) Evenness (J) (13):

$$J = \frac{\bar{H}}{\text{LnS}}$$

where $\bar{H}$ = Shannon-Weaver index of diversity based on numbers or weights
S = total number of species in sample
Ln = natural logarithm

(3) No/km: relative abundance measured as number of individuals caught per km electrofished

(4) Kg/km: relative abundance measured as aggregate weight of fish caught per km electrofished

(5) Composite Index of Well-Being (I_{wb}) (4, 5)

$$I_{wb} = 0.5 \text{ Ln(No/km)} + 0.5 \text{ Ln(kg/km)} + \bar{H}_{(no.)} + \bar{H}_{(wt.)}$$

where No/km = number individuals per kilometer electrofished
Kg/km = total weight per kilometer electrofished
$\bar{H}_{(no.)}$ = Shannon-Weaver index of diversity based on numbers
$\bar{H}_{(wt.)}$ = Shannon-Weaver index of diversity based on weights

Results

Sampling effort for the two streams was quite comparable with 63 samples

taken for a total sampling distance of 31.5 km from Big Vermilion River and 76 samples taken for 29.6 km from Sugar Creek. Forty species were collected from Big Vermilion River and 48 from Sugar Creek (Table 1). More fish were collected from Sugar Creek than from Big Vermilion River (2148 compared to 999), but they were much smaller on the average (10.2 g/individual compared to 35.6 g/individual), so that a much greater aggregate weight of fish was collected from Big Vermilion River.

TABLE 1: *List of fishes collected by electrofishing Big Vermillion River and Sugar Creek during 1979 and 1980.*

Family and Scientific Name	Common Name	Big Vermilion R.	Sugar Creek
Petromyzontidae (Lampreys)			
Ichthyomyzon unicuspis	Silver lamprey	X	
Lampetra appendix	Amer. brook lamprey		X
Acipenseridae (Sturgeons)			
Scaphirhynchus platorynchus	Shns. sturgeon	X	
Lepisosteidae (Gars)			
Lepisosteus osseus	Longnose gar	X	X
Lepisosteus platostomus	Shortnose gar	X	X
Amiidae (Bowfins)			
Amia calva	bowfin		X
Clupeidae (Herrings)			
Dorosoma cepedianum	Gizzard shad	X	X
Alosa chrysochloris	Sj. herring	X	X
Hiodontidae (Mooneyes)			
Hiodon tergisus	Mooneye	X	
Esocidae (Pikes)			
Esox americanus	Grass pickerel	X	X
Cyprinidae (Minnows)			
Campostoma anomalum	Stoneroller	X	X
Cyprinus carpio	Carp	X	X
Ericymba buccata	Silverjaw minnow	X	X
Hybopsis amblops	Bigeye chub	X	X
Hybopsis x-punctata	Gravel chub	X	X
Nocomis micropogon	River chub		X
Notropis blennius	River shiner	X	X
Notropis atherinoides	Emerald shiner	X	X
Notropis rubellus	Rosyface shiner	X	X
Notropis spilopterus	Spotfin shiner	X	X
Notropis stramineus	Sand shiner	X	X
Phenocobius mirabilis	Suckermouth minnow	X	X
Pimephales notatus	Bluntnose minnow	X	X
Semotilus atromaculatus	Creek club	X	X
Notropis chrysocephalus	Striped shiner	X	X
Notropis whipplii	Steelcolor shiner		X
Catostomidae (Suckers)			
Carpiodes carpio	River carpsucker	X	X
Carpiodes cyprinus	Quillback	X	X
Carpiodes verifer	Highfin carpsucker	X	X
Catostomus commersoni	White sucker		X
Hypentelium nigricans	No. hog sucker	X	X
Ictiobus bubalus	Smallmouth buffalo	X	
Minytrema melanops	Spotted sucker		X
Moxostoma anisurum	Silver redhorse	X	X
Moxostoma duquesnei	Black redhorse	X	X
Moxostoma erythrurum	Golden redhorse	X	X
Moxostoma macrolepidotum	Sh. redhorse	X	X
Ictaluridae (Catfishes)			
Ictalurus natalis	Yellow bullhead	X	X

TABLE 1.—Continued

Family and Scientific Name	Common Name	Big Vermilion R.	Sugar Creek
Ictalurus punctatus	Channel catfish	X	X
Noturus miurus	Brindled madtom	X	X
Pylodictis olivaris	Flathead catfish	X	X
Percichthyidae (Basses)			
Morone chrysops	White bass	X	X
Centrarchidae (Sunfishes)			
Ambloplites rupestris	Rock bass	X	X
Lepomis macrochirus	Bluegill	X	X
Lepomis megalotis	Longear sunfish	X	X
Micropterus dolomieui	Smallmouth bass	X	X
Micropterus punctulatus	Spotted bass	X	X
Pomoxis annularis	White crappie	X	X
Pomoxis nigromaculatus	Black crappie		X
Percidae (Perches)			
Etheostoma blennioides	Greenside darter	X	X
Percina caprodes	Logperch	X	X
Percina maculata	Blackside darter	X	X
Percina phoxocephala	Slenderhead darter	X	X
Percina sciera	Dusky darter	X	X
Stizostedion canadense	Sauger	X	
Stizostedion vitreum	Walleye		X
Sciaenidae (Drum)			
Aplodinotus grunniens	Freshwater drum	X	

Between 82% and 88% of the catches from both streams were made up of the same 14 species of fish (Table 2). Gizzard shad were equally abundant in both streams, but the relative abundance of most other species was quite different. Catches from Sugar Creek were dominated by hog suckers, black and golden redhorse, river chub, spotfin and striped shiners, longear sunfish, and smallmouth bass. Catches from Big Vermilion River were dominated by carp and northern river carpsucker.

TABLE 2: *Catch rates of the 14 most common species.*

Species	Sugar Creek		Big Vermilion R.	
	No/km	Kg/km	No/km	Kg/km
Gizzard shad	13.70	0.90	13.14	1.12
Spotfin shiner	12.69	0.07	1.24	0.01
Northern hog sucker	9.42	1.41	1.75	0.26
River club	6.85	0.23	0	0
Black redhorse	4.52	1.07	0.38	0.10
Striped shiner	3.34	0.06	0	0
Golden redhorse	3.21	0.82	1.91	0.71
Longear sunfish	2.87	0.14	1.68	0.09
Smallmouth bass	2.57	0.35	0.64	0.08
Stoneroller	2.06	0.03	0	0
Bluntnose minnow	1.59	0.01	0.67	0.01
Northern river carpsucker	0.03	0.02	3.02	2.27
Highfin carpsucker	0.10	0.03	0.32	0.15
Carp	0.44	0.87	2.73	5.16
Other species	9.10	1.40	4.23	1.32
Totals	72.49	7.39	31.71	11.28

Many more different species of Cyprinidae were collected from Sugar Creek than from Big Vermilion River, which may account in part for the much greater number of species taken per collection in the former stream (Table 3). Calculated diversities, evenness, and the composite index were also substantially higher in Sugar Creek.

TABLE 3: *Community statistics for electrofishing catches from Sugar Creek and Big Vermilion River (± 1 S.E.).*

Paramater	Sugar Creek	Big Vermilion R.
No./Km.	72.32(5.93)	31,71(3.35)
Kg/Km.	7.42(0.60)	11.28(1.72)
Ave. No. species/collection	8.22(0.44)	4.92(0.32)
Shannon diversity (no.)	1.67(0.06)	1.15(0.07)
Shannon diversity (wt.)	1.36(0.06)	0.87(0.07)
Evenness (no.)	0.84(0.01)	0.71(0.04)
Evenness (wt.)	0.67(0.02)	0.53(0.04)
Composite index	5.86(0.20)	4.30(0.26)

The data also provide a trophic analysis when each species of fish is assigned to a predominant feeding category on the basis of literature evidence (3, 16, 14, 1, 19). Based upon weight, the fish population of Big Vermilion River is dominated by omnivores (46.4%) and detritivores (22.6%), while Sugar Creek is dominated by insectivores (51.3%) (Table 4). A much greater proportion of the catch by weight consisted of piscivores, species of special interest to fishermen, in Sugar Creek (16.6%) than in Big Vermilion River (6.1%).

TABLE 4: *Trophic composition (%wt) of the electrofishing catches of fish from Sugar Creek and Big Vermilion R.*

Feeding Guild	Sugar Creek	Big Vermilion R.
Piscivores	16.6	6.1
Insectivores	51.3	14.9
Herbivores	12.6	9.9
Omnivores	17.7	46.4
Detritivores	1.7	22.6

Discussion

The combined influence of several factors accounts for the differences in the fish communities of Big Vermilion River and Sugar Creek. Big Vermilion River contains more species characteristic of large rivers, such as shovelnose sturgeon, shortnose gar, drum, and smallmouth buffalo, although at low flow the two streams are not that different in size. The lower gradient of Big Vermilion River leads to the development of long pools with short riffles scattered at intervals of one to three miles. The quality of habitat is generally much lower in Big Vermilion River than in Sugar Creek although for at least one-third of the collecting stations habitat quality was subjectively judged on a par with that of Sugar Creek.

These factors no doubt have some influence on the fish community of Big Vermilion River, but cannot entirely account for the very low composite index

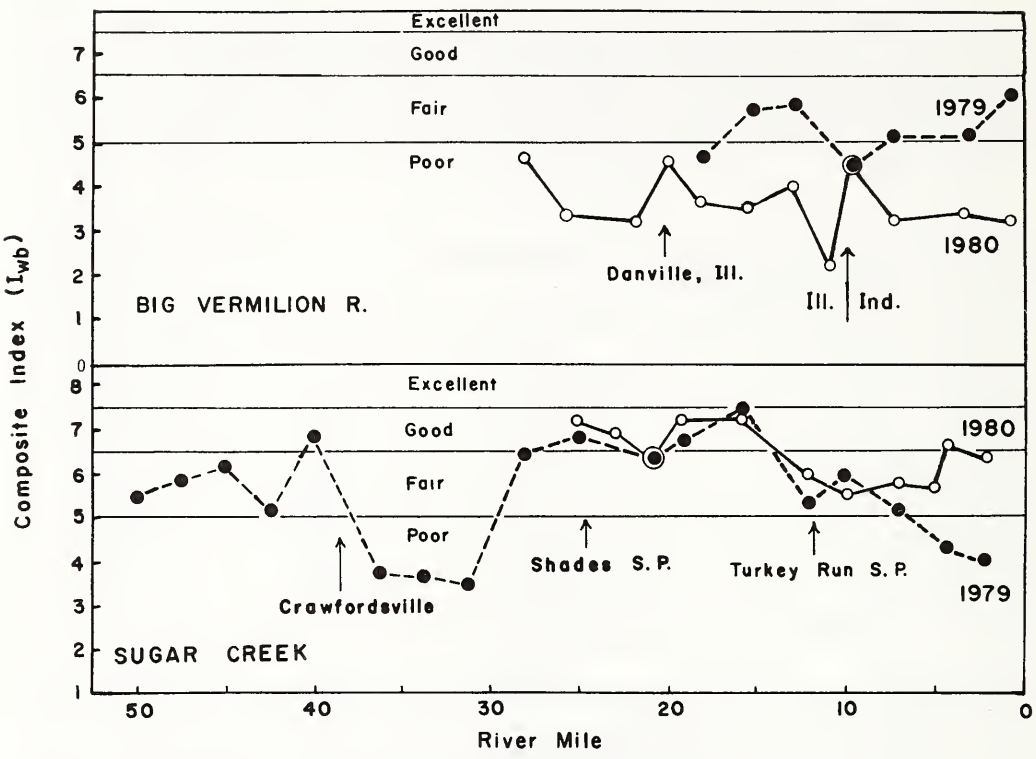

FIGURE 1: *Composite index profile for Big Vermilion River and Sugar Creek.*

values noted throughout the lower stream (Figure 1) which generally indicate poor to degraded environmental conditions (5). Even upstream from Danville, Ill. the fish community gives evidence of stress, probably the result of poor water quality from Salt Fork since habitat quality was excellent. In the Danville area itself, trash litters the river banks, industrial fumes cut through the woodland smells, old barren strip mines slope nearby, and a green scum of algae develops in slow-water regions during low-flow. Visual improvements occur downstream, but environmental quality and fish communities do not. There is no evidence that the river improves with distance from Danville. Perhaps any improvements in water quality are offset by the increased lateral erosion so strongly evident in the lower five miles.

In summary, the fish communities of lower Big Vermilion River are unworthy of the beautiful, wooded valley through which it flows. There is no doubt that the environmental degradation of the Big Vermilion River ultimately extends into the Wabash River into which it flows. Potentially the river is a valuable recreational asset, but the problems besetting its waters are widespread and complex and it is unrealistic to expect their solution in the near future.

Unlike Big Vermilion River, Sugar Creek is relatively insulated from the encroachment of man throughout most of its lower length, but the same problems afflict it to a less extensive degree. The best fish communities are located in 15 miles of stream in the Shades and Turkey Run State Park area, fish communities which are perhaps as good as any in the state of Indiana at this time in terms of population abundance, number of species, diversity and density of game species. Nearly 23% of the catch here consisted of gamefish species, with a particular abundance of smallmouth bass.

The communities are less good in the 10 miles of stream between Darlington and Crawfordsville, although since the 1979-80 collections improvements have been

made in the waste treatment process at Darlington. There was no indication that the municipal electric generating plant at Crawfordsville influenced the fish, but the stream was strongly degraded for 10 miles downstream from the Crawfordsville sewage treatment plant. After recovery has occurred in the vicinity of the state parks another zone of degradation occurs in the lower 15 miles of the stream largely because of agriculture.

Some tributaries in this lower section carry large quantities of silt and fertilizers into Sugar Creek. A notable example is a small creek which enters from the north just above the narrows near Turkey Run State Park which was usually choked by mud. In addition, as the river flows west from U.S. 41 it enters a broad shallow valley where fields extend up to the bank and lateral erosion is strongly evident. The depression of the fish communities from these entirely agricultural activities generally termed as nonpoint source pollution is fully as extensive as from the point source of Crawfordsville.

Sugar Creek is an outstanding recreational resource, providing pleasure for thousands of campers, canoeists, and fishermen. Recent improvements in waste treatment at the state parks and Darlington will help maintain the value of Sugar Creek. Improved waste treatment at Crawfordsville is needed and an extensive program aimed at reducing sheet and lateral erosion in the agricultural fields in the lower valley is also clearly indicated in the future to prevent further degradation. Probably the upper, eastern part of the watershed should be examined to assess problem areas here also. Overall, Sugar Creek is in fair to good environmental condition and every effort should be made to keep or, in places, improve its present quality.

Unfortunately, the impact of agriculture in the lower 10 or so miles means that Sugar Creek is not the positive factor it might be with respect to the environment of the Wabash River. Nevertheless, on at least one occasion this stream served as a vital refuge for fish trying to survive low dissolved oxygen concentrations in the Wabash River itself (6).

Acknowledgments

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Editor's Note: The various spellings (Vermilion/Vermillion) are in agreement with the different spellings used on both Indiana and Illinois maps.

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Phytoplankton Community Ordination at a Site in Extreme Southern Lake Michigan

THOMAS E. LAUER
Division of Water Pollution Control
Indiana State Board of Health
Indianapolis, Indiana 46206
and
THOMAS S. MCCOMISH,
Department of Biology
Ball State University
Muncie, Indiana 47306

Introduction

The phytoplankton community existing in the extreme southern basin of Lake Michigan near Michigan City, Indiana, was evaluated from June 1973 to May 1974 using principal components ordination analysis. This analysis is a mathematical technique that can reduce a large volume of data to a simplified objective presentation (10). Qualitative and quantitative phytoplankton data are used as "loading factors" to calculate ordinal placement values for each sample included in the analysis (7). A graphical display of the principal components can visually depict similarities and differences within the community. This exercise enabled temporal variabilities over the period of study, and the spatial limitations in this portion of Lake Michigan, to be evaluated. Although numerous studies investigating seasonal changes and spatial differences have been completed on Lake Michigan since 1872 (2,6,14,17,18,19,20), ordination analysis was typically not applied.

A number of community ordination techniques exist, each having a specific application (1,10,21). Orloci (10) notes that principal components is the most efficient of ordination techniques because it maximizes data point spread. Pielou (12) indicated it is the most straightforward of many possible techniques, but some authors note that mathematical calculations are more difficult than other methods (4,10). Although the limitations of principal components ordination technique have been noted (4,10,12), its application to community analysis is beneficial in interpretation of results.

Study Area

Eight stations were located in Lake Michigan on three transects near Michigan City, Indiana. Stations 1-4 were located 1770 m east of the Michigan City lighthouse at depths of 5, 10, 15, and 18 m, respectively, on Michigan City transect (M). Stations 5 and 6 were located at 5 and 15 m depths on the Dunes transect (D) 2800 m west of the lighthouse near the dune known as "Old Baldy." Stations 7 and 8 were located on a transect about 3470 m west of the harbor lighthouse near the mouth of Kintzele Ditch (K). McComish (9) described the transects and study area in detail.

Methods

Field and Laboratory

One-liter water samples for phytoplankton analysis were collected at about monthly intervals at stations in Lake Michigan from June 1973 to May 1974.

Samples were collected at the surface in wide mouth polypropylene bottles and preserved with merthiolate solution (23).

Plankton samples in the bottles were allowed to stand and settle in the laboratory for a minimum of two weeks. Samples were aspirated to about 40 ml and remaining settled plankton concentrate was placed into a 150 ml container. Aspirated supernatant was passed through a Foerst centrifuge to collect any organisms not settled out, and the centrifugate was then added to the sample concentrate. Samples were subsequently adjusted to standard 100 ml volume by adding preservative. Three 1 ml subsamples were taken from each sample concentrate with a Hensen-Stemple pipet and each was placed into a Sedgewick-Rafter counting chamber. Three strips representing 20% of the counting chamber volume were counted at 160X magnification. Non-diatoms were enumerated and identified to species when possible, while diatoms were counted and recorded as only pennate or centric for application of a proportional count. Permanent slides were prepared for indentification of diatoms in each sample following the procedure outlined by Weber (23). A total of about 500 diatoms were identified for each sample using a Zeiss Nomarski interference contrast microscope at 1000X magnification. The individual species tally was then applied to the proportional Sedgewick-Rafter cell count following Weber's (23) methods. Taxonomic keys for phytoplankton identification included Prescott (13), Patrick and Reimer (11), Weber (24), Drouet and Daily (3), Smith (16), and Tiffany and Britton (22).

TABLE 1. *Species used in the principle components ordination analysis*

Species
Bacillariophyta
Asterionella formosa
Cyclotella glomerata
Cyclotella meneghiniana
Cyclotella michiganiana
Cyclotella ocellata
Diatoma tenue var. *elongata*
Fragilaria crotonensis
Fragilaria intermedia
Melosira islandica
Melosira italica
Navicula cryptocephala
Navicula exigua
Nitzschia palea
Nitzschia sublinearis
Stephanodiscus astraea
Stephanodiscus binderanus
Stephanodiscus hantzschii
Stephanodiscus minutus
Stephanodiscus tenuis
Synedra ulna
Tabellaria fenestrata
Tabellaria flocculosa
Thalassiosira fluviatilis
Chlorophyta
Ankistrodesmus falcatus
Lagerheimia ciliata
Scenedesmus dimorphus
Scenedesmus quadricauda
Sphaerocystis schoeteri
Cyanophyta
Chroococcus limneticus

Ordination

Principal components ordination as described by Orloci (10) was employed in community evaluation of the phytoplankton assemblage. Since the analysis is not sensitive to small populations, and the intent of the ordination is to show major community direction (Ladewski, per. comm.) only "major" species were included in the analysis. The species selected occurred in at least 25 percent of the total samples and had a minimum population of 6/ml in a single sample. This eliminated 197 of the 226 taxa identified in the study. Loading factors, or input data, of these 29 species (Table 1) were based on percentage, not actual densities, in an effort to normalize the data from the 85 samples used.

The first three ordinal values were calculated, and displayed graphically, using the X, Y, and Z axes. Mathematical computations were completed at the University of Michigan Computing Center using programs supplied by the Great Lakes Reserach Division, the University of Michigan.

Results

Principal components ordination analysis using the first and second components shown on X and Y axes, respectively, revealed a clumping of stations, based primarily on temporal cycling (Figure 1). Three major seasonal groups were defined: June-September, October-January, and March-May. Only minor trends were noted for spatial differences. Each major season grouping had additional variability revealed by the third component, represented by the Z axis (Figure 1). Although not as significant as the first or second component values, in some instances the third component separated individual stations within a group. A relatively small number of species were responsible for the major groupings as well as influencing some of the lesser differences found within each defined season. Other

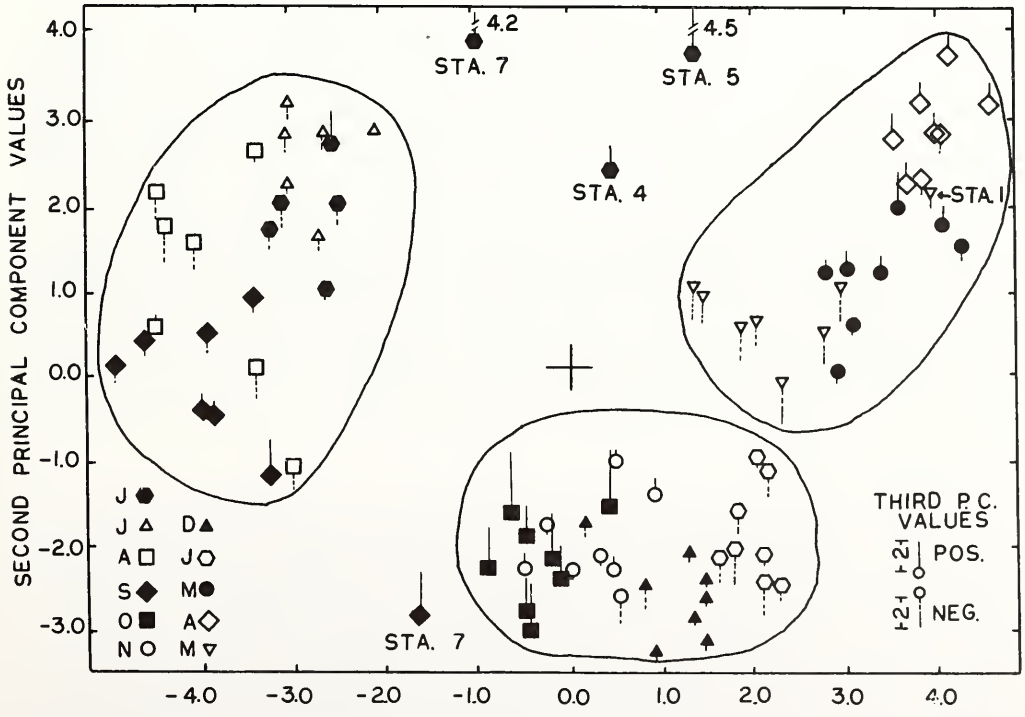

FIGURE 1. *First Principal Component Values*

less dominant species did not significantly affect definition of the community but could have accounted for some of the minor differences found between and within months.

June-December Grouping

The occurrence of *Thalassiosira fluviatilis* had the most profound effect on the ordination of the summer community. High populations of this diatom were found in June at stations 1, 2, 3, 6, and 8 with densities ranging from 25 to 74% of the total community. Absence of this species at stations 4, 5, and 7 was a primary factor causing ordinal displacement of these stations (Figure 1). Additionally, *Stephanodiscus binderanus, S. hantzschii, S. minutus and Ankistrodesmus falcatus* had distinctly higher populations at stations 4, 5, and 7 than the remaining five stations in June. *Cyclotella meneghiniana* and *Nitzschia palea* populations appeared notably higher at stations 1, 2, 3, 6 and 8 during this period, however. On the basis of both examinations of the raw data and the ordination analysis, two communities existed in June.

Thalassiosira fluviatilis and *Cyclotella glomerata* dominated all stations in July. Ordinal placement of stations during this period reflected similar population values for all samples. Continued dominance by *T. fluviatilis* in August ordinated these samples near earlier summer ones as expected. Minor changes during this time, however, were principally due to the absence of *C. glomerata* (a July dominant), and the strong appearance of *Chroococcus limneticus* and *Lagerheimia cilata.*

In September, *Thalassiosira fluviatilis* again dominated, but not at all stations. Station 7 showed relatively high populations of *Melosira italica, Fragilaria crotonensis* and *Tabellaria flocculosa.* Almost insignificant in this sample was *T. fluviatilis*, hence giving some explanation of the displacement of Station 7 away from the remaining September stations.

October-January Grouping

The two most important species influencing ordination of stations from October to January were *Tabellaria fenestrata* and *Fragilaria crotonensis.* These species dominated the community through all the winter months, ordinating them into the second major grouping. Individual sampling months, however, were still grouped close together, as indicated by third component values. The secondary dominants responsible for this in October were *Tabellaria flocculosa, Chroococcus limneticus* and *Melosira italica.* Changes in secondary dominants in November to *Asterionella formosa* and *Stephanodiscus minutus* displaced these samples slightly from the previous month. A large increase of *T. fenestrata* and *S. minutus* in December and January, and *F. intermedia* and *M. islandica* in January ordinated these samples somewhat away from October and November. The overall community structure during this four month period, however, was not markedly different.

March-May Grouping

A distinct change in dominance from pennate to centric diatoms resulted in a shifting of ordinal station placement in the samples. *Stephanodiscus minutus* was the dominant species in March, ranging from 16 to 33% density at the eight stations. *Melosira islandica, M. italica*, and *Stephanodiscus binderanus* were all present in high densities in March. April stations were characterized by a general decline of the four March dominants. Nine of the 29 species used in the ordination had average abundance levels above 5% during April, making the single most abundant species less important as an ordinating factor. The general trend for

May was a community more similar to winter months, with *Tabellaria fenestrata* and *Fragilaria crotonensis* increasing in density. The most important species in May, however, was *Melosira islandica*, which dominated the community. The combination of these dominant taxa influenced ordinal placement of the May stations between the winter and remaining spring months. The high density of *Stephanodiscus* at station 1 along with relative low abundance of *M. islandica* ordinated this station apart from the remaining May stations.

Discussion

The close proximity of sample stations in the study area explains the general lack of spatial variability observed. The Kintzele Ditch and Dunes transects are only 700 m apart. Although the Michigan City transect is somewhat separated from the other two, it is only about 4600 m northeast of the Dunes transect. The Michigan City Harbor area, including the influences of Trail Creek, the Northern Indiana Public Service Company generating station, and the high volume of boating and recreational activity, did not appear to greatly influence ordination of the phytoplankton communities considered in the study area. Spatial variability is usually only shown over a large expanse of the lake, or where significant environmental factors such as major rivers or point source wastewater inputs exist (15). This did not appear to be the case in this study. The occasional ordinal displacement of a single sample away from the remaining samples within a month was not consistent throughout the study. Sampling bias (5,8) or the plankton patchiness common in all aquatic systems (25) could explain some of these variations.

Although statistical significance could be found among individual stations for given populations in certain months, the general overview delineated by the principal components analysis revealed general community stability in the three major seasonal groupings. The year-long sampling regimen provided a good portrait of the temporal succession of species dominance. Minor changes occurred in principal component groups from June through September, from October through January, and March through May. Major shifts from one relatively stable period to the next occurred as the result of temporal shifts in species and abundance. The changeover of species dominance appears to be cyclic, but gaps appearing in the ordination may not have been as pronounced if sampling had been more frequent. This seasonal succession in Lake Michigan has been shown by several other investigators (6, 19, 20).

Principal components ordination analysis allowed graphic evaluation of relationships between samples, sites, and dates. Since the analysis was restricted to the numerically dominant taxa, it did not include all of the species collected. It did, however, express community similarities for an objective overview of the changes in phytoplankton community structure over the year.

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Seven Years of Forest Succession in Happy Valley, Jefferson County, Indiana

Christian J. Martin and Paul C. MacMillan
Department of Biology, Hanover College,
Hanover, Indiana 47243

In April, 1974, a tornado severely damaged the forest in Happy Valley, Jefferson Co., Indiana. Pre-tornado data indicate that the forest was a near-mature, second-growth, sugar maple-Ohio buckeye community. A tree census in 1974 after the tornado revealed the loss of about one-third of the trees, and that sugar maple, white ash and white oak spp. had best survived the perturbation.

Seven years after the storm the portion of the forest most severely damaged in 1974 is dominated by sugar maple, elm spp., Ohio buckeye and redbud, and the sugar maple, white ash, and white oak spp. remain dominant only in the least disturbed portion of the valley. Mean distance between trees in the storm-affected area was 4.4 m pre-tornado (1973), 4.9 m post-tornado (1974) and 3.3 in 1981. The mean number of trees per hectare was 511 pre-tornado, 425 post-tornado and 927 in 1981. These changes in tree population provide evidence of secondary succession in Happy Valley.

Introduction

On the afternoon of April 3, 1974, a major tornado travelled across portions of Jefferson Co. in southeastern Indiana, traversed diagonally across the wooded Happy Valley, and created extensive damage. In the fall of 1974, Bailey & MacMillan (1) completed a census of those standing live trees (i.e., dbh ≥ 5 cm) which remained. Their findings indicated that the storm reduced the tree population in the valley by as much as one-third. With substantial areas of the canopy removed and a portion of the forest floor freed from supporting mature trees, Happy Valley became a natural laboratory for studying secondary succession.

Analysis of pre-tornado data, collected at different times by Dan Webster (pers. comm.) and John Davis (pers. comm.), suggests that the valley forest was a sub-climax sugar maple-Ohio buckeye community. Following the tornado, Bailey and MacMillan (1) described the forest as a sugar maple-white ash-white oak spp. community. The objectives of this study were to (1) catalog the present composition of the forest, (2) describe any quantitative changes that have occurred in the seven years following the storm, and (3) hypothesize about further succession in the valley.

Study Site

The study site is located at 38° 43′ N and 85° 27′ W, on the Hanover College campus, Jefferson County, Indiana. It is an area of rather sharp relief; from a plateau 230 m above sea level the wooded slopes descend 75 m to a narrow valley floor. An intermittent stream flows through the valley and empties into the Ohio River. The portion of the valley under consideration is approximately one-half of its total length (Figure 1).

Three line transects were made across the valley; two transects lay completely within the windthrown area while the third lay substantially south and out of this zone (Figure 1). These three transects were the same as those measured by Bailey and MacMillan (1).

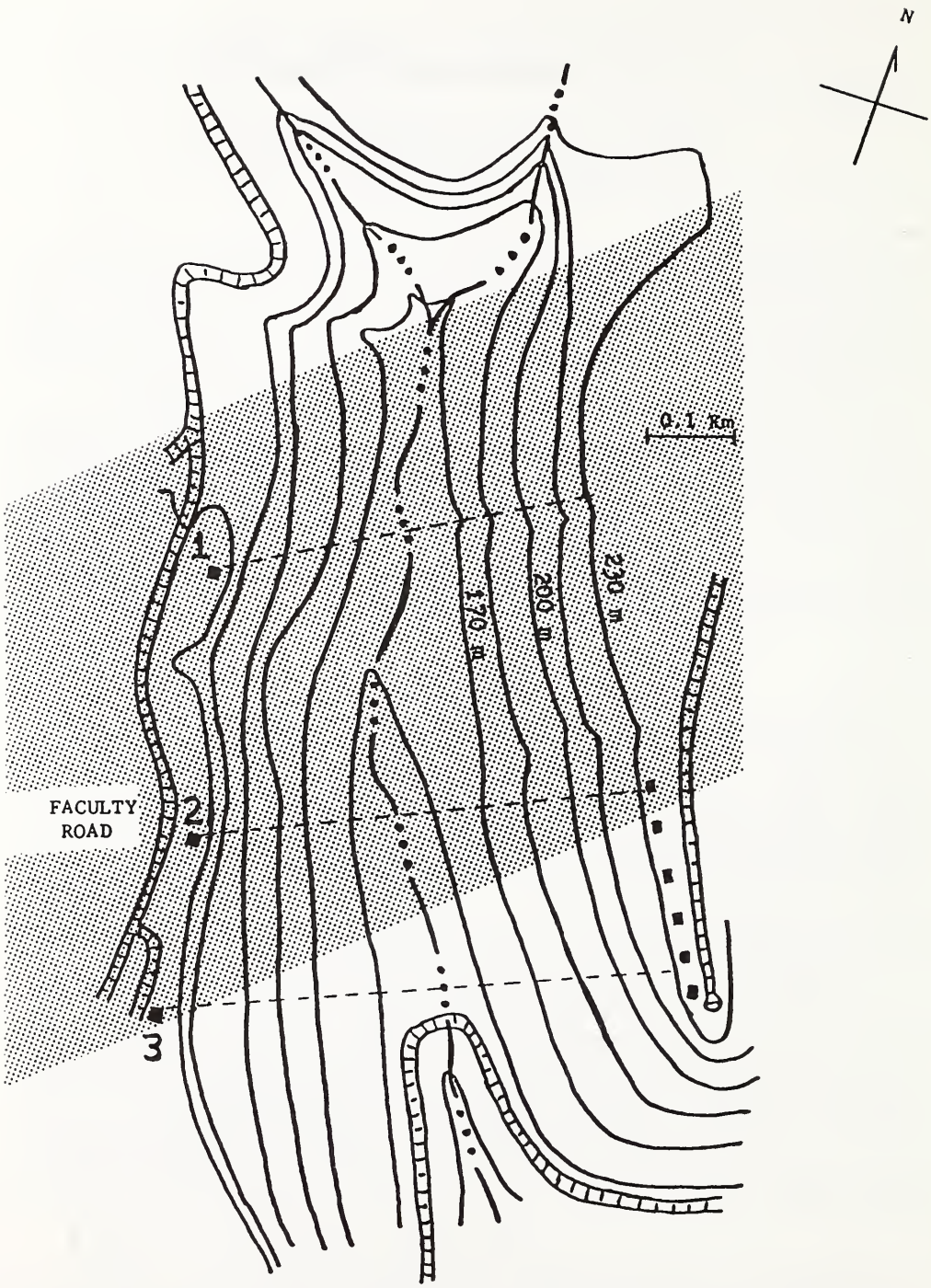

FIGURE 1. *Topographic map of Happy Valley study site. Numbers 1, 2 and 3 designate origins of the three transects. Contour intervals: 15 m. The shaded area represents the path of the tornado.*

Methods

Using the point-quarter method of Cottam and Curtis (3) with intervals of 15 m (50 ft), a random sample of the tree population was obtained. Data recorded in the field included point-to-tree distance, diameter at breast height (dbh), species

identification and estimated tree height (i.e., small = < 3 m, medium = 3 to 9 m, tall = > 9 m).

Field data were analyzed using the methods of Bailey and MacMillan (1) and Cox (4). These methods use transect data to estimate mean distance and mean area per tree, and numbers of trees per hectare. To facilitate comparisons, the data were divided into four size classes based on dbh. These classes were: Class 1 = 5 to 10 cm dbh, Class 2 = 11 to 20 cm dbh, Class 3 = 21 to 30 cm dbh, and Class 4 = > 31 cm dbh. Computations were then performed to estimate relative density (RD), relative frequency (RF), relative basal area (RBA), and relative importance value (RIV) of each species within each of the four size classes (1).

Transects 1 and 2 represent that portion of the study site most seriously damaged by the tornado; data from these two transects were combined to yield specific information about this area, and are referred to as "woods" data. In the "woods" summary relative density and relative frequency were computed in the same manner as above, but without reference to the size classification. In addition, the following computations were used in place of RBA and RIV: Relative Dominance (RDom) of a species and Importance value (IV) of a species (1).

Criteria for Inclusion in Sample

One procedural problem in this study was how to judge whether or not an individual tree should be included in the sample. Any major wind-storm, such as the 1974 tornado, will result in a population of maimed survivors which are exceptions to the normal morphological definition of a tree (i.e., having a single vertical axis, commonly exceeding 3 m in height and usually devoid of low branches, but bearing a head or crown of branches and leaves). The standard tree is relatively uncommon in Happy Valley, where uprooted, wind-snapped, leaning and bent boles have survived and adapted to compensate for the structural damage suffered in the storm. New trunks have arisen from living root systems and from what were secondary branches. In addition, many trees exhibit scars as a result of being struck, bent or splintered during the storm.

To gather an unbiased sample, we established the following criteria. A "tree" must: (a) be at least 5 cm dbh, (b) be no more than 45° from vertical at the point of measurement, (c) show evidence of being alive (i.e., fresh buds, flexible twigs), and (d) not be so severely damaged by recent events (i.e., storms and fallen trees) that its survival is in doubt.

Results

General data describing the past and present tree population in Happy Valley are summarized in Table 1. The present mean point-to-tree distance and mean area of each tree are much lower than post-tornado values, and lower than pre-tornado means. The number of trees per hectare has increased by 165% in Transect 1, 75% in Transect 2 and 12% in Transect 3, as compared to the census data taken immediately following the tornado (1).

Tables 2 through 4 contain summaries of Transects 1, 2, and 3, respectively, by size classes. Species are ranked according to relative importance values (RIV). Overall, sugar maple (*Acer saccharum*) is the single most important tree species in the valley; it has the greatest RIV in size classes 1, 2, and 3, except in Transect 1, Class 1, where it is second to slippery elm (*Ulmus rubra*).

In Transect 1, Class 1 (Table 2), sugar maple and American elm (*Ulmus americana*) showed marked decreases in RIV, while slippery elm, American

TABLE 1. *Pre-Tornado (1973), Post-Tornado (1974), and Present (1981) conditions of the Happy Valley Forest, Jefferson County, Indiana.*

	Mean Distance (m)	Mean Area (m2)	Trees Per Hectare
Pre-Tornado Summary (J. Davis, Pers. Comm.)	4.4	19.5	512
Post-Tornado Summary (Bailey & MacMillan, 1977)	4.9(A)	23.6(A)	425(A)
	3.9(B)	15.2(B)	652(B)
1981 Summary	3.3(A)	11.0(A)	927(A)
	3.7(B)	13.7(B)	730(B)

(A) Mean Data From Transects 1 and 2 Through the More Damaged Portion of the Forest.
(B) Data From Transect 3 Through the Less Damaged Portion of The Forest.

TABLE 2. *Transect 1, Class 2. The Following Symbols are used in Tables 2 through 4: RD = Relative Density; RF = Relative Frequency; RBA = Relative Basal Area; RIV = Relative Importance Value; An Asterick (*) Denotes a Species Datum Discussed in the Text; Numbers Enclosed by Parentheses Indicate the Rank Given to the Species in 1974 (1).*

Species	RD	RF	RBA	RIV
*Slipperty Elm (10)	21.7	18.7	24.1	64.5
*Sugar Maple (1)	13.0	14.6	15.8	43.4
*American Basswood (11)	15.9	12.5	14.6	43.0
*Redbud	14.5	12.5	11.5	38.5
Black Maple (3)	8.7	8.3	10.3	27.3
*Wild Black Cherry	5.8	6.2	4.7	16.7
White Ash (5)	4.3	6.2	3.4	13.9
American Elm (2)	4.3	4.2	5.3	13.8
Hop-Hornbeam (4)	2.9	4.2	3.1	10.2
Rock Elm	2.9	4.2	2.2	9.3
Ohio Buckeye (6)	1.4	2.1	1.9	5.4
White Oak Spp. (8)	1.4	2.1	1.2	4.7
Hackberry (9)	1.4	2.1	1.0	4.5
Blue Ash	1.4	2.1	0.8	4.3
Totals	99.6	100.0	99.9	299.5

TABLE 2A. *Transect 1, Class 2.*

Species	RD	RF	RBA	RIV
*Sugar Maple (1)	36.4	36.4	25.2	98.0
*Ohio Buckeye (2)	27.3	27.3	38.3	92.9
*Slippery Elm (4)	18.2	18.2	10.2	46.6
Black Maple (5)	9.1	9.1	13.5	31.7
*White Oak Spp. (3)	9.1	9.1	12.8	31.0
Totals	100.1	100.1	100.0	300.2

TABLE 2B. *Transect 1, Class 3.*

Species	RD	RF	RBA	RIV
*Sugar Maple (2)	36.4	36.4	29.5	102.3
*White Oak Spp. (1)	27.3	27.3	30.5	85.1
Ohio Buckeye	9.1	9.1	11.9	30.1
Blue Ash	9.1	9.1	11.9	30.1
American Elm	9.1	9.1	8.4	26.6
Black Maple	9.1	9.1	7.7	25.9
Totals	100.1	100.1	99.9	300.1

TABLE 2C. *Transect 1, Class 4.*

Species	RD	RF	RBA	RIV
Sycamore (2)	25.0	25.0	43.7	93.7
Red Oak Spp. (3)	25.0	25.0	30.3	80.3
American Elm	25.0	25.0	14.9	64.9
Sugar Maple	25.0	25.0	11.1	61.1
Totals	100.0	100.0	100.0	300.0

TABLE 3. *Transect 2, Class 1.*

Species	RD	RF	RBA	RIV
*Sugar Maple (1)	20.8	20.0	26.1	66.9
*Slippery Elm (2)	19.4	18.3	20.3	58.0
*Ohio Buckeye (4)	8.3	8.3	9.8	26.4
*Redbud (2)	8.3	8.3	7.1	23.7
Rock Elm	6.9	5.0	7.8	19.7
Black Maple	5.6	5.0	4.4	15.0
*American Basswood (6)	4.2	5.0	4.3	13.5
Hackberry (6)	4.2	5.0	3.9	13.1
Hop-Hornbeam	4.2	5.0	3.0	12.2
Royal Paulownia	2.8	3.3	2.2	8.3
Blue Ash	2.8	3.3	1.8	7.9
Box Elder	2.8	3.3	1.7	7.8
Tulip Tree	2.8	3.3	1.5	7.6
Papaw	2.8	1.6	1.7	1.6
Honey Locust	1.4	1.6	1.9	4.9
*White Ash	1.4	1.6	1.9	4.9
Butternut	1.4	1.6	0.7	3.7
Totals	100.1	99.5	100.1	299.7

TABLE 3A. *Transect 2, Class 2.*

Species	RD	RF	RBA	RIV
*Sugar Maple (1)	65.2	53.3	60.0	178.5
*Ohio Buckeye	26.1	33.4	31.5	91.0
American Elm	4.3	6.7	5.3	16.3
Black Maple	4.3	6.7	3.3	14.3
Totals	99.9	100.1	100.1	300.1

TABLE 3B. *Transect 2, Class 3.*

Species	RD	RF	RBA	RIV
*Sugar Maple (1)	53.3	50.0	51.6	154.9
*Hackberry (4)	13.3	14.3	17.4	45.0
*Ohio Buckeye (3)	13.3	14.3	10.1	37.7
Sycamore	6.7	7.1	8.2	22.0
American Elm (6)	6.7	7.1	7.3	21.1
White Oak Spp.	6.7	7.1	5.4	19.2
Totals	100.0	99.9	100.0	299.9

TABLE 3C. *Transect 2, Class 4.*

Species	RD	RF	RBA	RIV
Ohio Buckeye	100.0	100.0	100.0	300.0

TABLE 4. *Transect 3, Class 1.*

Species	RD	RF	RBA	RIV
*Sugar Maple (1)	28.0	26.2	27.4	81.6
*Slippery Elm (5)	14.0	9.5	13.0	36.5
*Papaw	10.5	9.5	9.0	29.0
*Hop-Hornbeam	7.0	9.5	5.6	22.1
*Hackberry (3)	5.3	4.8	7.1	17.2
*Box Elder (8)	7.0	4.8	4.8	16.6
*Ohio Buckeye (6)	5.3	4.8	6.4	16.5
*White Oak Spp. (9)	3.5	4.8	6.2	14.5
*White Ash (10)	3.5	4.8	5.4	13.7
*American Elm (4)	3.5	4.8	2.4	10.7
Wild Black Cherry	1.8	2.4	2.3	6.5
Black Maple	1.8	2.4	2.0	6.2
Red Mulberry	1.8	2.4	2.0	6.2
American Basswood	1.8	2.4	2.0	6.2
Kentucky Coffeetree	1.8	2.4	1.8	6.0
Black Walnut	1.8	2.4	1.5	5.7
Flowering Dogwood	1.8	2.4	0.9	5.1
Totals	100.2	100.3	99.8	300.3

TABLE 4A. *Transect 3, Class 2.*

Species	RD	RF	RBA	RIV
*Sugar Maple (1)	36.4	38.1	28.2	102.7
*Box Elder (5)	13.6	14.3	18.2	46.1
*White Ash (3)	13.6	14.3	13.0	40.9
*White Oak Spp. (2)	13.6	9.5	14.8	37.9
Black Maple (9)	9.1	9.5	8.8	27.4
Ohio Buckeye	4.5	4.8	6.6	15.9
*Slippery Elm (4)	4.5	4.8	5.4	14.7
Wild Black Cherry	4.5	4.8	5.1	14.4
Totals	99.8	100.1	100.1	300.0

TABLE 4B. *Transect 3, Class 3.*

Species	RD	RF	RBA	RIV
*Sugar Maple (1)	32.0	33.3	31.4	96.7
*White Ash (2)	16.0	19.0	18.9	53.9
*White Oak Spp. (3)	20.0	14.3	19.3	53.6
Hackberry (6)	16.0	19.0	13.9	48.9
Box Elder	8.0	4.8	8.0	20.8
American Elm	4.0	4.8	4.8	13.6
Black Maple (9)	4.0	4.8	3.7	12.5
Totals	100.0	100.0	100.0	300.0

TABLE 4C. *Transect 3, Class 4.*

Species	RD	RF	RBA	RIV
*Sycamore (3)	27.8	20.0	41.4	89.2
*American Elm (4)	11.1	13.3	22.0	46.4
Sugar Maple	16.7	20.0	7.9	44.6
*Black Walnut (2)	16.7	13.3	8.4	38.4
*Red Oak Spp.	5.6	6.7	9.1	21.4
Hackberry (7)	5.6	6.7	4.4	16.7
Bitternut Hickory (8)	5.6	6.7	2.4	14.7
White Ash	5.6	6.7	2.3	14.6
Box Elder (6)	5.6	6.7	2.0	14.3
Totals	100.3	100.1	99.9	300.3

basswood (*Tilia americana*), and redbud (*Cercis canadensis*) showed dramatic increases since 1974. In Transect 1, Class 2, sugar maple decreased in RIV, but remained more important than Ohio buckeye (*Aesculus glabra*) which increased. In Transect 1, Class 3 (Table 2b) sugar maple was less important that the various white oak spp. (*Quercus bicolor, Q. michauxii, Q. muehlenbergii, Q. prinus*) in 1974, however, in 1981, white oak spp. showed a considerable decrease.

In 1981 sugar maple, slippery elm, Ohio buckeye and redbud remained the dominant species in Transect 2, Class 1 (Table 3); however sugar maple RIV had decreased since 1974. White ash (*Fraxinus americana*), which had ranked fourth in 1974, was located only once in the 1981 census. In Transect 2, Class 2 (Table 3a), sugar maple remained most important, while Ohio buckeye, not found in 1974, increased in RIV to second in 1981. Sugar maple, Ohio buckeye and hackberry (*Celtis occidentalis*) remained most important in Transect 2, Class 3 (Table 3b).

For the area less damaged by the storm, Transect 3, Class 1 (Table 4), eight of the ten species with highest RIV's in 1981 were the same as in 1974. Four of the five most important species in Transect 3, Class 2 (Table 4a) in 1974 had the highest RIV's again in 1981; however, sugar maple showed a decrease. In Transect 3, Class 3 (Table 4b) sugar maple, white ash and white oak spp. maintained RIV rankings of first, second and third, respectively, but sugar maple exhibited a decrease in RIV. Two large American elms measuring 89 and 56 cm dbh, established this species as second most important in Transect 3, Class 4 (Table 4c). In addition, a single red oak sp., measuring 67.5 cm dbh, placed this species fifth in RIV, while sycamore (*Platanus occidentalis*), sugar maple and black walnut (*Juglans nigra*) ranked first, third and fourth, respectively.

Table 5 contains "woods" data (a summary of all classes on Transects 1 and 2) from the present study, with comparisons to 1974. The 1981 "woods" data showed that while decreasing in RIV, sugar maple remained the most important species. Increased RIV's for Ohio buckeye, slippery elm and redbud placed these species second, third and fourth in importance, respectively. White oak spp. and white ash, which were second and third in 1974, decreased in 1981.

Discussion

The damage done to the forest in Happy Valley by the tornado of 1974 resulted in the loss of ca. 80% of the canopy trees (1). However, some trees remained (even though damaged) which could have provided seeds, understory trees which had been suppressed were released and many damaged canopy trees resprouted to some extent. Thus, there were ample sources for revegetation of the forest, which should have led to a moderate rate of succession (2). Bailey and MacMillan (1) found that the north end (Transect 1) of the study site was hit hardest by the tornado and cited as evidence the increase in mean distance between trees and the mean area per tree, going from south to north (Figure 1 and Table 1) up the valley. In the present study, a complete reversal of this mean distance and mean area gradient was found; mean distance and mean area per tree now decrease from south to north through the study site (Table 1). Areas of the site most disturbed by the storm of 1974 are now the areas of greatest tree density, albeit due primarily to trees in size Class 1. Clearly, a growth of trees passing 5 cm dbh minimum is suggested.

If we use the criteria of change in mean distance and mean area as an indicator of succession, we can now suggest that the area of Transect 3 was also affected by the tornado because we found decreases in mean distance between trees and in mean area per tree and an increase in the number of trees per hec-

tare on this transect. These date substantiate the observation of downed trees in the area of Transect 3 after the tornado (pers. observ.).

Throughout the study site sugar maple continues to be the dominant tree. Stearns (5) suggested that sugar maple can maintain dominance in an area because of its ability to reproduce and grow successfully in shade as well as clearings. This doubtless accounts for its overall abundance in Happy Valley.

Transect 1 was extensively damaged in 1974 and the Class 1 trees (5 to 10 cm dbh) have greatly increased in density in the seven years since the tornado; this reflects both an increase in diversity and an increase in relative importance value (RIV) over those species present in 1974. Slippery elm and American basswood had RIV ranks of 10 and 11, respectively, in 1974, and redbud was not located during the 1974 census, but these three species are now three of the four most important species in this Transect and Class. According to Wharton and Barbour (7), slippery elm grows on dry slopes and moist, well-drained bottomlands. American basswood prefers well-drained soils; redbud is common on cut-over or wind-thrown areas in calcereous soils, while wild black cherry, also relatively important in this study area, becomes established readily in forest openings and is typical of the forest trees which regenerate rapidly when the vegetation is cleared (6). The Happy Valley study site, extensively damaged by the 1974 tornado, has provided prime habitat for all of these sun-tolerant, rapidly-growing trees.

Transect 1, Class 2 trees (11 to 20 cm dbh) were similar in 1974 and 1981. The same top five species were found and the order of imporatance remained much the same; however, rapid-growing Ohio buckeye increased its RIV ca. 100% while sugar maple decreased. In Transect 1, Class 3, sugar maple has replaced white oak spp. as the most important species due to a 37% decrease in RIV of white oak spp. In Transect 1, Class 4, only four trees > 31 cm dbh were found; thus all have large RIV's.

Transect 2 was also extensively damaged in the storm; since then growth of saplings into Class 1 has been rapid and species diversity has increased. White ash is an interesting exception; plentiful in 1974, it was much less important in 1981. A possible explanation is that a reduction in the supply of seeds has occurred in this part of the valley. During the course of study many large white ash boles were found, wind-thrown by the 1974 storm. These trees were once the primary source of white ash seeds along this transect, but in 1981 not one living white ash larger than 10 cm dbh was located along this transect. Combined with the better survival of sugar maple and slippery elm, a decrease in relative importance of white ash might be expected.

Transect 2, Classes 2, 3 and 4 showed little change in the seven years since the initial census by Bailey and MacMillan (1) due to the length of time required for smaller trees to grow into the larger size classes.

Transect 3 was least affected by the tornado, and relative stability is the most obvious characteristic of this transect. In Class 1, eight of the ten most important species were the same in 1974 and 1981. In Class 2, four of the five most important species in 1974 were also most important in 1981. In Class 3, the three most important species were the same in 1974 and 1981. However, in both classes 2 and 3 the RIV of slippery elm declined.

Transect 3, Class 4 contained several species which rated as relatively important because of their large diameter, rather than their abundance. Such a situation is typical of a more mature forest, which the southern end of the valley represents.

Table 5, "woods" data, represents a summary of that portion of the study site most heavily damaged by the tornado in 1974. Differences between the values

TABLE 5. *Summary of Data Collected From Transects 1 and 2, Through the Most Storm-Damaged Part of the Forest, Referred to as "Woods" Data. RDOM = Relative Dominance, Other Symbols the Same as in Tables 2 Through 4.*

Species	RD	RF	RDOM	RIV
*Sugar Maple (1)	27.1	22.3	32.1	81.5
*Ohio Buckeye (6)	10.1	12.8	11.7	34.6
*Slippery Elm (5)	15.0	14.9	4.2	34.1
*Redbud (14)	7.7	7.4	1.5	16.6
Black Maple (10)	6.3	6.1	3.7	16.1
*American Elm (8)	3.4	3.4	7.6	14.4
American Basswood (13)	6.8	6.1	1.5	14.4
Sycamore (4)	1.0	1.3	11.9	14.2
White Oak Spp. (2)	2.9	4.1	7.2	14.2
Hackberry (9)	2.9	4.1	4.4	11.4
Red Oak Spp.	0.5	0.7	7.0	8.2
Rock Elm	3.4	3.4	0.8	7.6
Blue Ash	1.9	2.7	2.2	6.8
Hop-Hornbeam	2.4	3.4	0.5	6.3
White Ash (3)	1.9	2.7	0.4	5.0
Wild Black Cherry	1.9	2.0	0.4	4.3
Royal Paulownia	1.0	1.3	0.2	2.5
Box Elder	1.0	1.3	0.1	2.4
Tulip Tree	1.0	1.3	0.1	2.4
Papaw	1.0	0.7	0.1	1.8
Honey Locust	0.5	0.7	0.2	1.4
Butternut	0.5	0.7	0.1	1.3
Totals	100.2	103.4	97.9	301.5

obtained by Bailey and MacMillan (1) and those obtained in 1981 indicate the trend of secondary succession here. In "woods" data, sugar maple remains most important although its RIV decreased by 33 since 1974. Ohio buckeye rose in RIV from sixth in 1974 to second in 1981; this change was not as dramatic in individual Transect/Class data (Tables 2 to 4). Thus, a definite trend toward restoration of Ohio buckeye as one of the dominant species in Happy Valley, as it was before the tornado (J. Davis, pers. comm.), is suggested. Slippery elm, which increased in importance in wind-thrown regions of the valley, appears to be entering a period of greater abundance. But judging from its disappearance from several of the larger diameter size classes since 1974, the ability of slippery elm to maintain its increased importance over the long-term is questionable. Redbud, which has increased vigorously since 1974 and now ranks fourth in importance, will probably decline in importance as taller species begin to close the canopy in this tornado-damaged portion of the valley.

In terms of secondary forest succession, the interval of time which has elapsed since Happy Valley was disturbed is quite short. Much of the windthrown region is still relatively open, and the forest canopy has not closed; however, the dramatic increase in trees per hectare (Table 1) in Transects 1 and 2, due almost entirely to saplings growing past the 5 cm dbh threshold, provides the first indication of a regenerating forest. Over the same period of time, the southern portion of the valley has changed much less. The area sampled by Transect 3 can still be described as a sugar maple-white ash-white oak spp. community, as it was by Bailey and MacMillan (1). However, the central and northern portions of the valley, sampled by Transects 1 and 2, no longer fit this description; the succession which has occurred since 1974 has resulted in what may best be described as a sugar maple-Ohio buckeye-slippery elm-redbud community. We expect that secondary

succession will continue to alter some of the dominant species of this forest community. The data discussed reveals some changes which have occurred in Happy Valley over a relatively short period of time.

Acknowledgments

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Full Tally Inventory of 4^+ cm Woody Individuals in Bendix Woods Nature Preserve, an Old Growth Beech-Maple Forest

VICTOR RIEMENSCHNEIDER
Department of Biological Sciences
Indiana University at South Bend
South Bend, Indiana 46634
and
THOMAS D. BLODGETT
Bendix Woods County Park
New Carlisle, Indiana 46552

Introduction

Bendix Woods County Park is located about 19 km west of South Bend, Indiana in western St. Joseph County. The Bendix Woods Nature Preserve is in the southwestern portion of the park and lies in the south half of section 11, T. 37 N., R. 3 E. (86° 29′ 16″. Longitude and 41° 39′ 11″ N. Latitude). The land containing the present park and preserve was homesteaded in 1860 by Jacob and Emily Hooton. In 1926, the portion of the land that includes the park was sold to the Studebaker Corporation and they constructed an automobile testing area that included several miles of roads. Later the Bendix Corporation purchased the testing grounds from Studebaker and in 1966, deeded 175 acres to the newly created St. Joseph County Parks and Recreation Board. During the last five decades, the preserve area has not been disturbed.

The park area is in the Kame facies of the Lagro Formation of the Wisconsinan glacial period (4). The elevation within the preserve ranges from 274 m to 242 m above sea level with slopes ranging from 2 to 22 percent. The soils are coarse textured (sandy loam), well drained members of the Hillsdale series (1). However, finer textured lenses do occur and several seasonal springs occur in the eastern third of the preserve which includes some moderately well to somewhat poorly drained soils.

The presettlement vegetation of the north one half of this township, as recorded by the surveyers in 1829, was dominated by sugar maple and beech with basswood, elm and white oak being major associates. The trees recorded for the 37 points in the north half were 28 percent sugar maple (avg. diameter, 54.8 cm) and 25 percent beech (avg. diameter, 44.5 cm). The largest beech and sugar maple recorded were 102 cm in diameter and the largest tree recorded was a 152 cm diameter walnut.

On July 5 and 9, 1980, straight line winds reaching velocities of more than 96 km per hr caused extensive damage in SW Michigan and northern Indiana. In the Nature Preserve, the wind impacted about 2.4 hectares or almost 25 percent of the preserve area. The present study was initiated in the spring of 1981 to assess the damage by the windstorm, to inventory the living woody individuals and to establish permanent quadrats to study forest regeneration. Tree, shrub and herbaceous level studies were initiated, but only the tree inventory has been completed and this should serve as baseline data for future studies.

Methods

A 50 X 50 meter grid was established for the preserve area using the SW corner of the preserve (corner post of chainlink fence) as the initial point. The

intersections of the 50 m interval, north-south and east-west lines were marked by driving 1.9 cm diameter by 61 cm long galvanized pipe into the ground. These corners of the permanent quadrats were established by measuring the sides and diagonals using 50 m and 100 m tapes. Elevation changes were measured and surface measurements corrected to establish a horizontal grid.

All woody stems within the inventory area (Figure 1) were tallied by subdividing the 0.25 ha permanent quadrats into 10 m X 10 m plots. The data recorded for each plot included the plot number, plot size and shape, date and the species, dbh and vegetation level for each woody stem, 4 cm or more in diameter. The plot number consists of the column and row numbers for the grid (Figure 1) plus the column and row of plot within quadrat. Shape and size of plot were recorded since the irregular boundaries of the preserve resulted in some rectangular and triangular plots. The field data were transcribed to a computer data bank for storage and analysis.

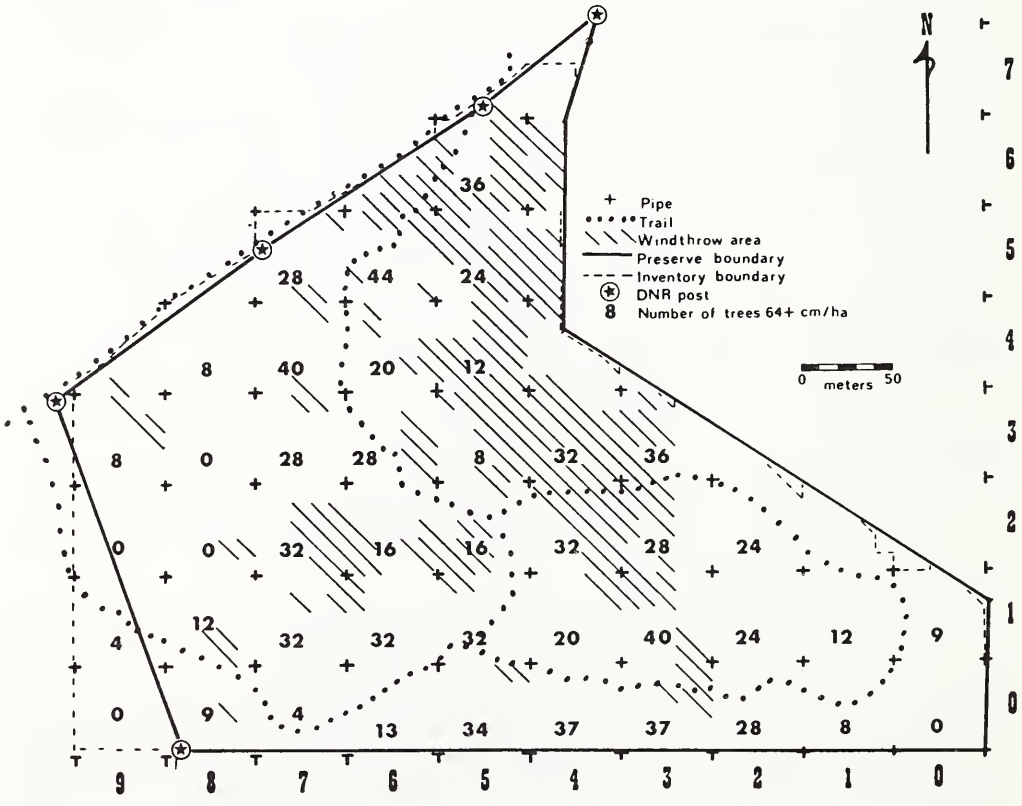

FIGURE 1. *Bendix Woods Nature Preserve and area included in full tally inventory of 11.6 ha in 1981 and 1982. Preserve is located in southeastern portion of Bendix Woods County Park.*

Results and Discussion

The summary statistics for the 11.6 ha inventoried are presented in tables 1 and 2. The inventory required more than one growing season to complete and the major windthrow area was the last to be inventoried. Therefore, a number of individuals may have entered the lower size class during the inventory period. The smallest size class is dominated by maple (*Acer saccharum* and *A. nigrum*), 73 percent of all individuals, and maple probably gained the most individuals through growth during the interval from windthrow to completion of inventory.

This dominance by maple in the lower size class skews the density and relative density values in table 1 and inflates the maple importance value.

TABLE 1. *Vegetation attributes for full tally inventory of woody individuals 4 cm or more dbh in Bendix Woods Nature Preserve, St. Joseph County, Indiana.*

	Vegetation attributes (2)				
Species (1)	D1	D2	B1	B2	IV
Acer saccharum	358.7	56.4	7.2	27.1	42
Fagus grandifolia	56.6	8.4	7.8	29.3	19
Ulmus rubra	56.7	8.9	3.7	13.9	11
Tilia americana	25.9	4.1	2.1	7.9	6
Liriodendron tulipifera	8.6	1.4	1.6	6.0	4
Ulmus americana	28.7	4.5	0.9	3.2	4
Celtis occidentalis	26.0	4.1	0.7	2.7	3.5
Carya cordiformis	21.1	3.3	0.7	2.7	3
Others	54.1	8.9	1.9	7.2	
TOTALS	636.4		26.7		

1. *Acer saccharum* includes *A. nigrum.* Other species in the 11.6 hectares inventoried area: *Asimina triloba, Carpinus caroliniana, Crataegus* sp., *Fraxinus americana, F. quadrangulata, F. pennsylvania, Gymnocladus dioica, Juglans cinerea, J. nigra, Ostrya virginiana, Platanus occidentalis, Populus deltoides, Prunus serotina, P. virginiana, Pyrus malus, Quercus rubra,* and *Sassafras albidum.*

2. Vegetation attributes are: D1, density in number per ha; D2, relative density in percent; B1, basal area in square meters per ha; B2, relative basal area in percent; IV, importance value, (D2 + B2)/2.

If the two species of maple (*A.saccharum* and *A.nigrum*) are separated, their importance values would be about the same as beech (*Fagus grandifolia*). If only canopy individuals were considered, the separate maple importance values would be less than beech and only slightly higher than slippery elm (*Ulmus rubra*). The forest would be classified as a beech-sugar maple-black maple community using the Indiana Heritage Program classification (2). The data for the two maple species are combined in this report since many individuals appear to be hybrids and no species distinction was made for about 80 individuals recorded in late fall and very early spring.

Most species in Table 1 have a similar rank for both the density and basal area values. Tuliptree (*Liriodendron tulipifera*) is a notable exception since it has a relatively low density value compared to its basal area value. Tuliptree has relatively few individuals in the smaller diameter classes and a relatively large number in the 40 to 60 cm classes (Table 2). A similar situation occurs in the frequency distributions of black cherry (*Prunus serotina*) and black walnut (*Juglans nigra*). Basswood (*Tilia americana*) has two secondary peaks in the larger size classes, beech has one and maple has a distict plateau in the 40 to 70 cm classes.

We believe past disturbances, particularly windthrow, have been important in shaping the observed frequency distributions. During the inventory, we noted much evidence of earlier windthrows throughout the preserve although none as massive as the present. The eastern and western two columns of quadrats (Figure 1) are much younger (smaller diameters) and probably reflect past cropping or logging of these areas. Black cherry is more common in the eastern and western portions of the preserve and tuliptree is more abundant in the western end. The eastern end is the lowest and wettest area of the preserve and contains the most massive of old windthrows. This end of the preserve also contains most of the young *Ulmus americana.* The heterogeneity of the preserve area explains some of the frequency variation in table 2. We hope to gain additional information on past disturbances through analysis of the growth rings of windthrown trees.

TABLE 2. *Frequency analysis of full tally data for Bendix Woods Nature Preserve, St. Joseph County, Indiana.*

Species (1)	Diameter classes (cm)								
	4 to	10 to 20	to 30	to 40	to 50	to 60	to 70	to 80	+
Acesp	3110	693	121	45	56	54	56	26	5
Carco	69	87	56	24	6	1	2	0	0
Celoc	160	91	30	7	3	4	1	4	2
Faggr	194	52	44	43	80	84	68	45	13
Frasp	49	55	44	22	10	0	0	0	0
Jugni	0	3	5	3	1	3	3	0	2
Lirtu	6	9	13	8	22	19	13	6	4
Pruse	2	16	11	23	14	0	2	0	0
Queru	1	4	7	7	0	1	2	0	2
Tilam	113	79	23	16	23	9	24	12	2
Ulmam	113	133	53	21	7	5	0	0	1
Ulmru	90	159	209	118	47	16	12	6	2
TOTAL	4246	1400	620	324	270	197	184	99	33
% *Acer*	73	50	20	13	21	27	30	26	15

1. Acesp, *Acer nigrum* and *A. saccharum*; Carco, *Carya cordiformis*; Celoc, *Celtis occidentalis*; Faggr, *Fagus grandifolia*; Frasp, *Fraxinus americana* and *F. pennsylvanica*; Jugni, *Juglans nigra*; Lirtu, *Liriodendron tulipifera*; Pruse, *Prunus serotina*; Queru, *Quercus rubra*; Tilam, *Tilia americana*; Ulman, *Ulmus americana*; Ulmru, *Ulmus rubra*. TOTAL includes all species in inventoried area, 11.6 ha.

In 1969, Lindsey et al. (3) reported a full tally inventory of individuals 10 cm or more dbh in 4.3 ha of Bendix Woods Nature Preserve. Although we are not sure of the exact location of the early survey, we think it was within the area defined by columns 4 through 7 and rows 1 through 5 in Figure 1. Table 3 is a comparison of the values for this 4.8 ha area and the data from Lindsey et al. The basal area values for the present inventory are slighly lower than Lindsey's data for beech, slippery elm and basswood. Since this area includes a portion of the major windthrow damage and these three species are represented by windthrown individuals, we believe the decrease in basal area reflects the amount of windthrow damage from the 1980 storms. In Figure 1, the windthrow damaged quadrats have fewer large trees and much lower basal area values than

TABLE 3. *Vegetation attributes for 1981 and 1969 inventories of 10+ cm trees in central old growth section of Bendix Woods Nature Preserve.*

Species (2)	Vegetation attributes (1)									
	D1		D2		B1		B2		IV	
Faggr	57.4	(69.7)	36	(38)	12.8	(13.9)	50	(49)	43	(43)
Acesp	51.5	(44.8)	32	(25)	5.0	(5.0)	19	(17)	26	(21)
Ulmru	21.8	(27.4)	14	(15)	2.7	(3.2)	10	(12)	12	(13)
Tilam	10.7	(13.7)	7	(7)	2.6	(3.0)	10	(10)	8	(9)
Celoc	9.0	(9.4)	6	(5)	0.8	(0.8)	3	(3)	4	(4)
Ulman	3.2	(5.0)	2	(3)	0.3	(0.5)	1	(2)	2	(2)
Lirtu	1.7	(3.0)	1	(2)	0.6	(0.6)	2	(2)	2	(2)
Carco	1.9		1		0.3		1		1	
TOTAL	161.1	(182.3)			25.7	(28.4)				

1. Vegetation attributes are: D1, density in number per ha; D2, relative density in percent; B1, basal area in square meters per ha; B2, relative basal area in percent; IV, importance value, (D2 + B2)/2. Data in parentheses are from Lindsey et al (3) for 4.3 ha. The other data are for 4.8 ha.
2. See table 2 for species codes.

the surrounding undisturbed quadrats. An inventory of the windthrown trees has not been completed so exact figures for loss of basal area are not available.

The basal area of the old growth portion of the preserve is comparable to that reported for other beech-maple forests in Indiana e.g. Hoot Woods, 25.2 m^2/ha, Officers Woods, 25.7 m^2/ha, Potzger Woods, 26.8 m^2/ha and Meltzer Woods, 28.8 m^2/ha (3). However, the 25.7 m^2/ha for the old growth portion is considerably less than the 39.6 m^2/ha reported for Warren Woods in Berrien County, Michigan (5). The largest tree in the Nature Preserve is a 105 cm beech. Warren Woods has several trees in this and larger size classes (5).

Although the beech and maple basal areas in Bendix Woods are similar to that of other Indiana beech-maple forests, none of the other forests had *Ulmus rubra* and *Tilia americana* as the major canopy associates. White ash (*Fraxinus americana*) and tuliptree are more important in the other forests than they are in Bendix Woods.

Summary

The central area of Bendix Woods Nature Preserve is a good example of an old growth beech-sugar maple-black maple forest with slippery elm and basswood as strong associates. The July 1980 windstorm damaged a large portion of the preserve and probably reduced the average basal area per hectare by 10 percent or more.

The establishment of permanent quadrats and the completion of a full tally inventory of the total preserve area provide the reference points and baseline data necessary to study the recovery from the 1980 windthrow and to assess future disturbances.

Acknowledgments

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Impingement of Fishes at the Indianapolis Power and Light Company Generating Station on the White River at Petersburg, Indiana

John O. Whitaker, Jr.
Department of Life Sciences
Indiana State University
Terre Haute, Indiana 47809

Introduction

Impingement is the lodging of organisms on the protective screens at the inflow channels of electric generating plants. The screens function to keep unwanted items out of the mechanism of the plant, and are automatically cleaned periodically by water running through them. The "impinged" items were previously allowed to move out a trough and back into the river, but now are removed. Impingement has caused concern as a possible hazard to fish at generating plants.

This work was part of a study of the effects of the Indianapolis Power and Light Company generating station on the White River at Petersburg, Indiana (Proffitt, 1969; Proffitt and Benda, 1971; Whitaker and Schlueter, 1973; Whitaker, Schlueter and Tieben, 1977). The objective of this phase of the work was to determine the extent of impingement of fish at the generating plant.

Materials and Methods

To determine effects of impingement on fish of the White River at the IPALCO plant, seines were placed across the impingement outflow trough and all fish were collected during extended sampling periods. The first seine was a 1/4-in. nylon mesh, and the second was of 1/8-in. nylon mesh. Both were heavily weighted. The nets were in position for 25-35 hours per collecting trip, and were checked periodically. Accumulated debris was removed and examined for the presence of fish, mollusks, and other large animal life. Early in the study, the fish were identified and numbers of the various species were simply counted, while later, they were assessed by size class. Also, during the latter part of the study, notation was made as to whether the fish were dead, alive, or in various stages of decomposition when collected. It was assumed that the "fresh" fish had been alive when impinged, but that the rotting fish had simply floated into the plant entrance after death. However, some of the fresh fish may have been dead when impinged, as dead fish can remain in fresh condition for several days in cold weather. On January 23-25, 1976, a sample was taken before, during and after deicing, to see if that process had any notable effect on the fishes impinged.

Eight reptiles were taken in the impingement sample, but are not included in the Tables. They included four map turtles, *Graptemys geographica*, three softshelled turtles, *Trionyx spinifer*, and one water snake, *Nerodia sipedon*.

Results

The total number, number per hour, and projected number per year of all species of fish taken in the impingement sample (excluding the deicing sample) are given in Table 1. These totaled 10,772 individuals. Of these, 9784 (90.8%) were gizzard shad, *Dorosoma cepedianum*, followed by *Ictalurus punctatus*, channel catfish (444, 4.1%) and *Ictiobus* sp., buffalo (149, 1.4%; young individuals of *I. niger* and *I. bubalus*). A total of 32 separate species is listed, along with some individuals identified only to genus and some unidentified fish.

TABLE 1. *Numbers of fish impinged during the 1399.75 hours (58.3 days) of sampling time at the IPALCO generating plant at Petersburg, together with number per hour and projected number per year.*

Species	Total No.	No/hr	Projected No/Year
Dorosoma cepedianum—Gizzard Shad	9784	6.99	61231
Ictalurus punctatus—Channel Catfish	444	0.32	2779
Ictiobus sp.—Buffalo	149	0.11	932
Aplodinotus grunniens—Freshwater drum	72	0.05	451
Pimephales vigilax—Bullhead minnow	46	0.03	288
Hybognathus nuchalis—Silvery minnow	35	0.03	219
Cyprinus carpio—Carp	30	0.02	188
Notropis spilopterus—Spotfin shiner	29	0.02	181
Ichthyomyzon unicuspis—Silver lamprey	26	0.02	163
Lepomis macrochirus—Bluegill	20	0.01	125
Pylodictus olivaris—Flathead catfish	19	0.01	119
Carpiodes carpio—River carpsucker	16	0.01	100
Hiodon alosoides—Goldeye	14	0.01	88
Lepomis cyanellus—Green sunfish	13	0.01	81
Micropterus punctulatus—Spotted bass	8	0.01	50
Pomoxis annularis—White crappie	6	0.004	38
Carpiodes velifer—Highfin carpsucker	6	0.004	38
Morone chrysops—White bass	6	0.004	38
Alosa chrysochloris—Skipjack herring	5	0.004	31
Lepomis megalotis—Longear sunfish	4	0.003	25
Stizostedion canadense—Sauger	3	0.002	19
Lepisosteus osseus—Longnose gar	3	0.002	19
Moxostoma sp.—Redhorse sp.	3	0.002	19
Hybopsis storeriana—Silver chub	3	0.002	19
Anguilla rostrata—American eel	1	0.001	6
Polyodon spathula—Paddlefish	1	0.001	6
Lepisosteus platostomus—Shortnose gar	1	0.001	6
Lepomis sp.—Sunfish sp.	1	0.001	6
Carpiodes cyprinus—Quillback	1	0.001	6
Scaphirhynchus platorhynchus—Shovelnose sturgeon	1	0.001	6
Carassius auratus—Goldfish	1	0.001	6
Ictiobus cyprinellus—Bigmouth buffalo	1	0.001	6
Semotilus atromaculatus—Creek chub	1	0.001	6
Ictiobus bubalus—Smallmouth buffalo	1	0.001	6
Ictalurus melas—Black bullhead	1	0.001	6
Unidentified fish	17	0.01	106
Totals	10,772	7.70	67,413

Seasonal distribution of the fish is given in Table 2. It is apparent that relatively few fish are generally impinged. Sampling periods usually varied from 25 to 35 hours, and the total number of fish taken was less than 100 individuals in all but 8 of the 23 sampling periods. The largest impingement collections occurred on September 13-14, September 20-21, September 27-28, and October 11-12, 1975, when totals of 404, 7432, 875, and 476 individuals were taken. Again, the great majority of these fish, totalling 8727 of the 9097 (95.9%) fish, were gizzard shad, *Dorosoma cepedianum*. These fish were mostly in the 2-4″ size classes, and thus included schools of young individuals

Tables 1 and 2 include all fish taken by impingement. However, some of the fish taken in these samples were in various stages of decomposition, and thus presumably were dead before drifting through the plant intake. Table 3 gives data for live and fresh fish in the later impingement samples. The total number of fish impinged during these periods was 10,533 in 928.5 hours, or 11.37 per hour.

TABLE 2. *Seasonal distribution of fish taken by impingement sampling at IPALCO generating plant at Petersburg, Indiana. Species in which at least 50 individuals were taken are summarized separately; rest are combined.*

	Numbers of fish							
Sampling Dates	*Dorosoma* cepedianum	*Ictalurus* punctatus	*Ictiobus* sp.	*Aplodinotus* grunniens	Other species	Total Fish	Total hrs.	No. fish per hr.
1973-74	95	6	0	0	17	118	181	0.65
1975								
Jan 25-27	1	0	0	0	2	3	46	0.06
June 13-15	13	0	0	0	5	18	52.25	0.34
28-30	6	0	0	0	2	8	51.5	0.16
July 11-13	1	7	0	2	4	14	52	0.27
18-20	6	13	0	0	4	23	45	0.51
25-27	30	17	0	1	7	55	46.5	1.18
Aug 3-5	4	4	2	0	5	15	48.5	0.31
9-11	11	42	2	0	27	82	49	1.67
13-15	11	115	3	16	17	162	52	3.12
23-24	11	6	4	1	4	26	28	0.93
30-31	12	18	3	2	3	38	38	1.00
Sept 6-7	7	6	5	0	4	22	33	0.67
13-14	156	117	90	7	34	404	31	13.03
20-21	7265	27	23	3	24	7342	31.75	231.24
27-28	842	16	3	1	13	875	31.75	27.56
Oct. 4-5	9	0	2	0	3	14	32	0.44
11-12	464	1	0	0	11	476	32	14.88
18-19	90	1	1	0	0	92	32	2.88
25-26	42	1	1	0	3	47	31.75	1.48
Nov 1-2	195	4	0	0	2	201	32	6.28
8-9	39	4	0	0	3	46	32	1.44
15-16	341	5	3	8	4	361	32	11.28
22-23	22	1	0	11	2	36	32	1.13
Dec 21-22	16	1	0	0	1	18	30	0.60
1976								
Mar 20-21	31	14	7	8	7	67	31	2.16
Apr 24-25	11	0	0	1	8	20	32	0.63
May 1-2	14	6	0	5	14	39	46	0.85
May 8-9	7	1	0	1	12	21	31.25	0.67
15-16	7	4	0	3	26	40	32	1.25
21-22	6	2	0	0	22	30	32	0.94
28-29	2	3	0	1	7	13	32	0.41
June 5-6	11	2	0	1	7	21	32	0.66
12-13	6	0	0	0	19	25	31.5	0.79
	9784	444	149	72	323	10,772	1399.75	7.70

Of these, 3767 were partially decomposed, leaving a total of 6,766 live or fresh fish, or 7.29 per hour. Again, the few samples with large numbers of small *Dorosoma* (Fall, 1975) contribute greatly to the overall calculated numbers per hour. In the 27 samples, there were 16 samples with less than 1 live or freshly killed fish per hour, and 21 with less than 3.

Most of the fish taken in the impingement samples were relatively small, less than 6 in. total length (many were 2-4″ long). Table 4 summarizes data for all fish over 6 in. long taken in the impingement sample from August 23, 1975,

TABLE 3. *Fish in varying stages of decomposition (1976), during 928.5 hours of sampling.*

1975	No. of fish	Total # dead and decaying	Hrs.	Total # of fresh fish	No/hr of live or freshly killed fish
Aug 3-5	15	10	48.5	5	0.10
9-11	82	80	49	2	0.04
13-15	162	58	52	104	2.00
23-24	26	25	28	1	0.04
30-31	38	35	38	3	0.08
Sept 6-7	22	14	33	8	0.24
13-14	404	184	31	220	7.10
20-21	7342	2989	31.75	4353	137.10
27-28	875	217	31.75	658	20.72
Oct 4-5	14	0	32	14	0.44
11-12	476	18	32	458	14.31
18-19	92	0	32	92	2.88
25-26	47	41	31.75	6	0.19
Nov 1-2	201	37	32	164	5.13
8-9	46	12	32	34	1.06
15-16	361	0	32	361	11.28
22-23	36	0	32	36	1.13
Dec 21-22	18	0	30	18	0.60
1976					
Mar 20-21	67	0	31	67	2.16
Apr 24-25	20	0	32	20	0.63
May 1-2	39	0	46	39	0.85
8-9	21	0	31.25	21	0.67
15-16	40	5	32	35	1.09
21-22	30	1	32	29	0.91
28-29	13	9	32	4	0.13
June 5-6	21	14	32	7	0.22
12-13	25	18	31.4	7	0.22
	10533	3767	928.50	6766	7.29

TABLE 4. *Size classes of fish impinged at the IPALCO generating plant at Petersburg, Indiana. (Most of the 10,274 fish taken between August 23, 1975, and June 1976 were less than 6 inches long. Fish greater than 6 inches total length are indicated here.)*

	Size groupings			
Species	7-12″	13-18″	18+ ″	Totals
Dorosoma cepedianum	51	0	0	51
Ichthyomyzon unicuspis	20	0	0	20
Hiodon alosoides	14	0	0	14
Cyprinus carpio	2	7	3	12
Ictalurus punctatus	6	0	0	6
Carpiodes carpio	4	0	0	4
Aplodinotus grunniens	3	1	0	4
Alosa chrysochloris	3	0	0	3
Moxostoma sp.	1	1	0	2
Scaphyrhynchus platorhynchus	0	0	1 (24″)	1
Morone chrysops	0	1	0	1
Pomoxis annularis	1	0	0	1
Ictiobus bubalus	0	1	0	1
Lepomis macrochirus	1	0	0	1
Ictalurus melas	1	0	0	1
	107	11	4	122

through June, 1976 (when length of impinged fish were recorded). Of the 10,274 fish taken during this period, only 122 or 1.2% of the individuals were longer than 6 in. Of these, nearly half were gizzard shad, and 20 were silver lampreys.

Another question of interest related to the effect of the deicing process (warming of the intake screens by spraying warm water into the intake just ahead of the screens) on the impingement of the fish. A combination of scheduling problems, equipment problems at the plant, and warm weather reduced the sampling when deicing was occurring to one period. This period was on January 23 to 25, 1976 (Table 5). On January 24, ambient river temperature was 4°C at the sur-

TABLE 5. *Fish taken by impingement sampling before and during deicing, Jan. 23-25, 1976, at IPALCO plant at Petersburg, Indiana.*

	21 hours *Before deicing*		*30 hours* *During deicing*		*51 hours* *Total*	
	No.	No/hr	No.	No/hr	No.	No/hr
Ictiobus sp.	21	1.0	46	1.53	67	1.31
Aplodinotus grunniens	12	0.57	10	0.33	22	0.43
Dorosoma cepedianum	2	0.10	16	0.53	18	0.35
Notropis atherinoides	2	0.10	0	—	2	0.04
Hybognathus nuchalis	1	0.05	0	—	1	0.02
Ictalurus punctatus	1	0.05	11	0.37	12	0.24
Morone chrysops	0	—	1	0.03	1	0.02
Lepomis macrochirus	0	—	1	0.03	1	0.02
Lepisosteus osseus	0	—	1	0.03	1	0.02
Totals	39	1.86	86	2.87	125	2.45

face. The impingement sampling seines were in place for 21 hours January 23-24 before deicing occurred. A total of 39 fish was taken, or 1.86 per hour. The deicers were then turned on and impingement sampling occurred for 30 additional hours and 86 fish were taken, or 2.87 per hour. This difference was not great, but was significant ($X^2 = 5.16$, 1 df).

It is unfortunate that more sampling periods were not possible in 1976 when deicers were operating. Additional data are needed, but it does appear that the deicer may attract some fish that otherwise would not have been impinged.

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ENGINEERING

Chairman: WARREN W. BOWDEN
Department of Engineering, Rose-Hulman Institute of Technology, Terre Haute, Indiana 47803

Chairman-Elect: DAVID D. CHESAK
Department of Physics, Saint Joseph's College, Rennsalaer, Indiana 47978

ABSTRACT

Measurement of Concentration Fluctuations in a Moving Stream. JEFFREY G. HIPPLER and THOMAS R. HANLEY, Department of Chemical Engineering, Rose-Hulman Institute of Technology, Terre Haute, Indiana 47803.—A conductivity cell and electronic signal processing circuit have been designed and built to measure the variance of concentration fluctuations in the exit stream of a mixed flow reactor. Experimental tests have shown that the variance measurements from a step input to a reactor were reproducible to within ten per cent, with a signal to noise ratio of greater than 100. Tests at different feed rates and agitation speeds in a mixed flow reactor produced a linear relationship between the logarithm of the concentration fluctuation variance and the reciprocal of the agitation level.

Antecedent Moisture Condition Probabilities for Selected Indiana Stations

Christopher B. Burke
School of Civil Engineering
Purdue University
West Lafayette, Indiana 47907
and
Donald D. Gray
Department of Civil Engineering
West Virginia University
Morgantown, West Virginia 26506

Introduction

The prediction of the volume of direct runoff resulting from a particular storm rainfall is among the central issues in hydrology. Abstractions such as interception, depression storage, and soil moisture storage cause direct runoff to be less than storm rainfall. The complexity and intrawatershed variability of these abstraction mechanisms has led to the adoption of simplified descriptions in many practically important runoff models. But even the most elementary models usually incorporate the observation that the direct runoff from a given storm normally increases with increasing moisture content of the watershed at the start of the storm.

A common scheme to account for the dependence of abstractions on initial watershed moisture is to define several discrete Antecedent Moisture Condition (AMC) classes based on the total precipitation during a specified period of time preceding the storm of interest. The model parameters which determine the abstractions are varied depending on the AMC class. It is easy to apply such schemes when calculating the runoff from historical storms if the appropriate antecedent rainfall data are available. To treat hypothetical or design storms, the analyst must postulate appropriate AMC classes, considering both the purpose of the calculations and the local climate. This paper provides a rational basis for such choices in Indiana by reporting the actual percentage of time spent in the various AMC clases of two widely used runoff models: the Soil Conservation Service Curve Number method and the Illinois Urban Drainage Area Simulator (ILLUDAS).

The Curve Number (CN) method is a simple empirical technique developed by the Soil Conservation Service (SCS) to calculate the direct runoff volume caused by a given depth of rain (12,14). The CN method applies to both rural and urban watersheds (15) and has been used around the world (7). It has been recommended for the calculation of excess rainfall in several unit hydrograph techniques (4, 8, 10, 13, 15). Hawkins (7) has provided a critical discussion of the theoretical basis of the CN method.

The heart of the CN method is the SCS Runoff Equation, a simple algebraic formula relating the direct runoff volume to the total rainfall volume and the Curve Number. The Curve Number reflects the hydrologic abstractions of the watershed. Using data from field experiments, the SCS has determined CN values for many combinations of soil type and artificial or natural cover (12, 15). These tabulated CN values are for an intermediate Antecedent Moisture Condition (AMC II). They must be decreased for drier soil (AMC I) or increased for wetter soil

(AMC III) in accordance with an empirical conversion table provided by the SCS (12, 15).

ILLUDAS is a non-proprietary Fortran program developed by the Illinois State Water Survey (18) to aid in the design or evaluation of storm sewer systems. Given the layout of an existing or proposed storm sewer network and an arbitrary rainfall hyetograph, the program calculates direct runoff hydrographs at each junction and specifies the necessary commercial pipe diameters. ILLUDAS is widely used in urban hydrology and its performance has been studied by the present authors (1, 2, 3) as well as by others.

ILLUDAS determines the direct runoff from grass covered areas by comparing the intensity of rain at each time step to the infiltration capacity predicted by Horton's empirical equation (9). Among the parameters in this equation are the initial and ultimate infiltration capacities. Different initial and ultimate infiltration capacities are specified for each of the 4 SCS soil types. The effects of antecedent moisture are modeled by defining 4 AMC classes with corresponding initial infiltration capacities for each soil type. A higher AMC class implies a wetter soil with a reduced initial infiltration capacity.

In the CN method and in ILLUDAS, the AMC for a given day is determined by the total rainfall on the 5 preceeding days. Table 1 lists the class limits used in this study. The dormant and growing season criteria for SCS AMC I, II, and III were established by the SCS (12). AMC IV was defined by the authors to quantify the percentage of time for which the CN method is invalid because the ground is frozen or snow covered. The originators of ILLUDAS established the limits for AMC 1, 2, 3, and 4 (18). AMC 5 was added by the authors. In the documentation for ILLUDAS, no distinction is made between the dormant and growing seasons. The authors believe that the infiltration capacities used in ILLUDAS are not valid during the dormant season in Indiana. For this reason the ILLUDAS criteria were applied only during the growing season. The operational definition of the growing season is discussed in the next section.

TABLE 1. *Antecedent Moisture Condition Criteria.*

AMC CLASS	TOTAL 5-DAY ANTECEDENT RAINFALL (INCHES)	
	Growing Season (April 1 - October 31)	Dormant Season (November 1 - March 31)
SCS AMC I	less than 1.4	less than 0.5
SCS AMC II	1.4 to 2.1	0.5 to 1.1
SCS AMC III	over 2.1	over 1.1
SCS AMC IV	Frozen soil or snow cover*	Frozen soil or snow cover*
ILLUDAS AMC 1	0	
ILLUDAS AMC 2	0 to 0.5	
ILLUDAS AMC 3	0.5 to 1.0	NOT
ILLUDAS AMC 4	over 1.0	APPLICABLE
ILLUDAS AMC 5	Frozen soil or snow cover*	

*The frozen condition is assumed to occur whenever the average daily air temperature is less than 32° F or whenever there is measurable snow on the ground.

Data and Analysis

This study was undertaken to provide Hoosier hydrologists and drainage engineers with occurrence probability estimates for SCS and ILLUDAS AMC classes. To accomplish this it was necessary to analyze long series of reliable daily rainfall and temperature observations from stations throughout the state. After consultation with State Climatologist Dr. Lawrence A. Schaal, the 10 stations listed in Table 2 were selected for analysis.

TABLE 2. *Seasonal AMC Occurrence Probabilities Expressed as Percentages.*

		SCS Growing				SCS Dormant				ILLUDAS				
Station	County	I	II	III	IV	I	II	III	IV	1	2	3	4	5
Berne	Adams	88	8	4	0	27	10	13	50	20	39	20	21	0
Mt. Vernon	Posey	86	7	6	0	35	13	19	33	27	34	16	23	0
Paoli	Orange	85	8	6	0	30	14	20	37	24	34	18	24	0
Rockville	Park	86	8	6	0	33	11	14	41	26	34	18	22	0
Rushville	Rush	87	7	5	1	29	10	14	47	25	34	19	21	1
Spencer	Owen	85	8	6	0	29	12	16	43	23	36	19	22	0
Valparaiso	Porter	85	8	6	0	23	8	11	57	19	38	20	22	0
Vevay	Switzerland	86	8	6	0	37	15	16	32	23	38	17	22	0
Waterloo	Dekalb	89	6	4	1	23	10	11	57	25	38	19	18	1
West Lafayette	Tippecanoe	86	7	6	1	27	9	10	54	22	37	18	22	1
Indiana average		86	8	6	0	29	11	14	46	23	36	18	22	0

Entries may not total 100% due to rounding.

Two magnetic tapes which contain all of the daily weather observations for every station in Indiana through 1977 were purchased from the National Oceanic and Atmospheric Administration. The daily record for each station used here includes the water equivalent precipitation, maximum and minimum air temperatures, and the depth of snow on the ground at the time of observation. These were the data used to determine the AMC for each day.

The most straightforward way to determine if the ground was frozen would be from actual soil temperature measurements, but these are available at only 12 sites in Indiana and begin in 1960 (16). Among the 10 stations selected for this study, only West Lafayette reports such measurements. In addition, soil temperature depends strongly on the depth of measurement and on the soil type. For these reasons it was not practical to use soil temperature to establish the occurrence of SCS AMC IV and ILLUDAS AMC 5.

McGarrahan and Dale (11) have investigated the accuracy of regression equations for the prediction of soil temperatures from air temperatures, but their results did not seem to warrant incorporating such a procedure in this work. Therefore, for the purpose of this study, "frozen ground" was deemed to occur if the average of the maximum and minimum air temperatures was less than 32.0°F. If this occurred or if there was more than a "trace" of snow on the ground at the time of the observation, the day was counted in SCS AMC IV and ILLUDAS AMC 5. The water equivalent of all precipitation falling during a series of "frozen ground" days (regardless of length) was accumulated and counted as rain on the last day of the series. This treatment neglects the possibility of gradual evapora-

tion or melting, but it is no more arbitrary than any other scheme to account for snowfall using the available data.

A Fortran program was written which read the appropriate data, formed 5 day antecedent water equivalent precipitation totals, and assigned each day to the appropriate AMC class. Whenever any of the 4 observations was missing, that day and the next 5 days were not assigned to an AMC class. The ratio of the number of days in each class to the total provides an estimate of the probability of the occurrence of that class.

The definition of the dormant and growing seasons required judgement. Climatologists often define the growing season as the period from the last air temperature of 32°F in the spring until the first air temperature of 32°F in the fall. To implement such a definition would require checking all the minimum temperatures until the first fall freeze before the season could be defined. Since each year's growing season is then different, it would be impossible to state whether growing or dormant criteria should be used during the spring and fall. These problems could be overcome by using average growing seasons, but these differ among stations. Average growing seasons have been established for 7 of the stations used in this study and for stations close to the others (17). The average date of the last occurrence of 32°F in the spring ranged from April 9 to May 5 with a mean date of April 25. The average date of the first fall occurrence of 32°F ranged from October 9 to October 25 with a mean date of October 17. To avoid the inconvenience of splitting months between the seasons, the growing season was defined to last from April 1 through October 31 for every station in this study.

The selection of a period of analysis for any climatological study requires a compromise among the need for a long period to assure statistical significance, the requirement that the period be short enough to reduce the influence of climatic change, and the desire for a short period to reduce the computational expense. The entire record for Berne from 1910 through 1977 was analyzed, and it was found that the 30 year period used for most climatological calculations was satisfactory for AMC studies as well. The results for 7 of the stations in this study are based on the period from January 1, 1948, through December 31, 1977. The exceptions are Spencer (1950-1977), Vevay (1961-1977), and West Lafayette (1954-1977), which began reporting at later dates. The percentage of days which were not assigned an AMC due to incomplete data ranged from 13% at Paoli to 1% at West Lafayette. The average was 7% with most such days occurring in the dormant season. The total number of days assigned to an AMC class was 94116.

Results

The primary results of this study are the seasonal AMC probabilities presented in Figure 1 and Table 2 for each station and for the state. The Indiana average entries are not simple averages of the station probabilities but are based on the total number of days analyzed.

The SCS growing season probabilities are remarkable for their uniformity among stations and for the overwhelming predominance of AMC I caused by the high class limit of 1.4 inches of rain. The probability that any growing season day will be in AMC I is everywhere greater than 85%. It is clearly incorrect to consider AMC II as an average condition. SCS AMC IV has a probability of less than 0.4% during the growing season, as does ILLUDAS AMC 5.

The SCS dormant season results show far more variation among stations, especially in AMC IV, the frozen soil or snow cover class. This probability ranges

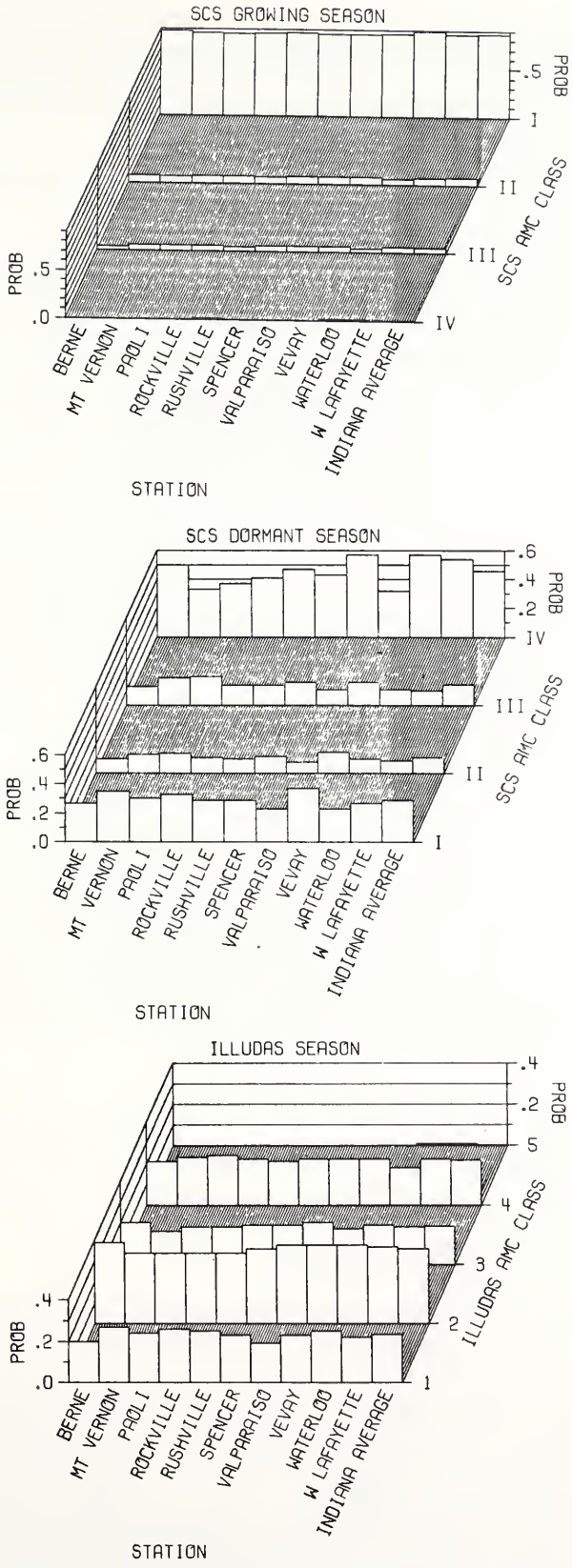

FIGURE 1. *Seasonal AMC Occurrence Probability Histograms for Each Station and for Indiana.*

from 32% at Vevay to 57% at Waterloo and Valparaiso. The trend correlates very closely with annual average temperature and with latitude, the colder stations having a greater AMC IV probability. The most important conclusion, however, is that the CN method is invalid for almost half of the dormant season for the state as a whole.

The ILLUDAS results, which refer to the growing season, show uniformity among stations and a fairly even distribution among classes. This makes the selection of a design AMC rather critical.

For some applications, monthly AMC probabilities are useful. Once again, the variations among stations are modest, except for SCS AMC IV during the dormant season which shows a marked increase with latitude. The Indiana average results are shown in Figure 2. The SCS growing season results vary slightly, with the probability of AMC I increasing from 84% in April to 90% in October. The AMC III probabilty ranges from 4% in October to 8% in June. The probability of AMC IV (and ILLUDAS AMC 5) is 1.7% in April, 0.4% in October, and 0% in the other growing season months.

During the dormant months, the probability of SCS AMC IV shows a definite seasonal trend with a maximum of 67% in January and a minimum of 19% in November. This variation occurs largely at the expense of AMC I, whose probability is least in January (15%) and greatest in November (23%).

The ILLUDAS probabilities show moderate variations. The probability of AMC 1 grows from a low of 13% in April to a high of 31% in October. The AMC 4 probability is greatest in June (27%) and least in October (15%).

Conclusions

The analysis of a total of 258 years of daily weather observations at 10 Indiana stations has provided reliable estimates of the probability of occurrence of the Antecedent Moisture Condition classes of the SCS Curve Number method and the ILLUDAS computer program. The uniformity of the probabilities across the state means that probabilities for any location in Indiana can be interpolated with confidence. The present results confirm and supercede the less statistically significant AMC probabilities published previously by Gray and his co-workers (5,6) for fewer Indiana stations.

Acknowledgment

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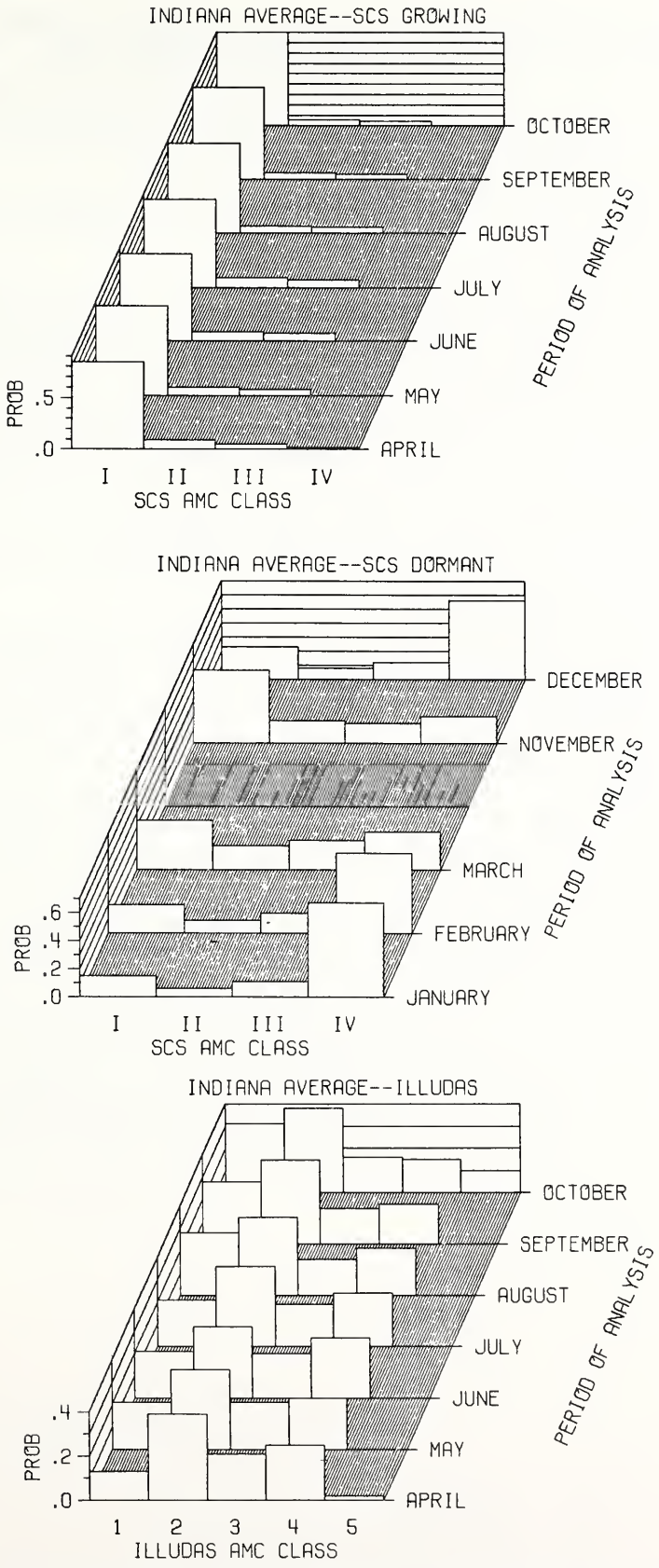

FIGURE 2. *Indiana Average AMC Occurrence Probability Histograms by Months.*

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Control and Measurement of Low Oxygen Partial Pressures in H_2 or N_2 Gas Using Calcia-Stabilized Zirconia Cells

KRIENGSAK CHALERMTIRAGOOL and WILLIAM E. STANCHINA
Department of Electrical Engineering
University of Notre Dame, Notre Dame, Indiana 46556

Introduction

Calcia-stabilized zirconia (CSZ) cells have been designed and fabricated to operate as an oxygen gauge to measure oxygen partial pressures in a gas flow (H_2 or N_2), and as an oxygen pump to increase or decrease oxygen partial pressures in it. In the atmosphere of H_2 flow, oxygen incorporation in it usually takes the form of $H_2O(g)$, whereas in N_2 oxygen can exist either as $O_2(g)$ or $H_2O(g)$ since there is a trace amount of H_2 in commercially supplied N_2. Control and measurement of low oxygen partial pressures were carried out over varying flowrates and pump currents in both flowing H_2 and N_2 gas. The results were presented and discussed for both gases.

Cell Operation

Calcia-stabilized zirconia (CSZ) is commercially available in various sizes, shapes and doping concentrations. Impurity doping causes defects in pure zirconia (ZrO_2) and is responsible for electrical (mainly ionic) conductivity. When pure zirconia is doped with divalent metal oxide such as calcium oxide (CaO), oxygen vacancies are created, one for each Ca atom. These doubly charged oxygen vacancies ($\dot{V}_O^{\cdot}$) are formed to maintain electrical neutrality with substitutional calcium. In the substitutional defect, a Ca^{2+} ion occupies a nominal Zr^{4+} ion site (represented by Ca_{Zr}) and has a relative charge of -2 [3]. When gases with different oxygen activities are present at the two opposite sides of the CSZ electrolyte and the temperature is raised sufficiently high, diffusion of oxygen ions will take place due to an oxygen concentration gradient across the electrolyte. This results in a reverse electric field which develops across the electrolyte in order to maintain equilibrium. The electric field can be equivalently described as being caused by the migration of oxygen vacancies in the direction opposite to that of oxygen ions until equilibrium is reached. Although electron diffusion is in general present, essentially all charge conduction is ionic at a cell operating temperature of 500°C. With one gas having a known amount of oxygen content, the cell emf represents a measure of the oxygen content in the other gas according to the relation [1,5]:

$$E = \frac{-RT}{4F} \ln \left[\frac{P_{O_2}}{P_{O_2}(\text{ref})} \right] \tag{1}$$

In H_2 gas, equation (1) can be modified to [1]:

$$E = \frac{-RT}{2F} \ln \left[\frac{(P_{H_2O}/P_{H_2})}{(P_{H_2O}/P_{H_2})\ \text{ref}} \right] \tag{2}$$

where

R = gas constant

T = cell temperature in K

F = Faraday equivalent

$P_{O_2(ref)}$ = reference oxygen partial pressure

$(P_{H_2O}/P_{H_2})ref$ = reference ratio of water partial pressure to hydrogen partial pressure

P_{O_2} = unknown oxygen partial pressure

$\frac{P_{H_2O}}{P_{H_2}}$ = ratio of unknown partial pressure of water to hydrogen partial pressure

When an electrical potential is applied across the cell, oxidation and reduction reactions will take place at the two electrolyte surfaces (electrodes) depending on the polarity of the applied potential. Oxygen is released at one electrode and captured at the other as a result of oxidation and reduction respectively. Thus a CSZ cell can be used as a pump to pump oxygen from one gas to the other through the electrolyte.

A CSZ gauge and a CSZ pump were fabricated using a 6″ long by 1/4″ O.D. and a 12″ long by 1/4″ O.D., respectively, slip cast tube made of ZrO_2 with 7.5 w/o CaO (Figures 1 and 2). Cell temperature was kept constant at 500°C by Eurotherm model 931 and 913 temperature controllers. The component layout and flow path of the gas flow system is illustrated in detail in Figure 3.

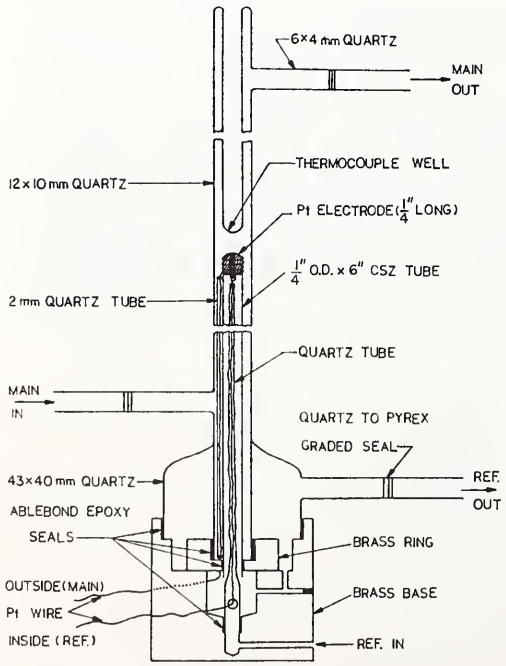


FIGURE 1. *CSZ Gauge Design*

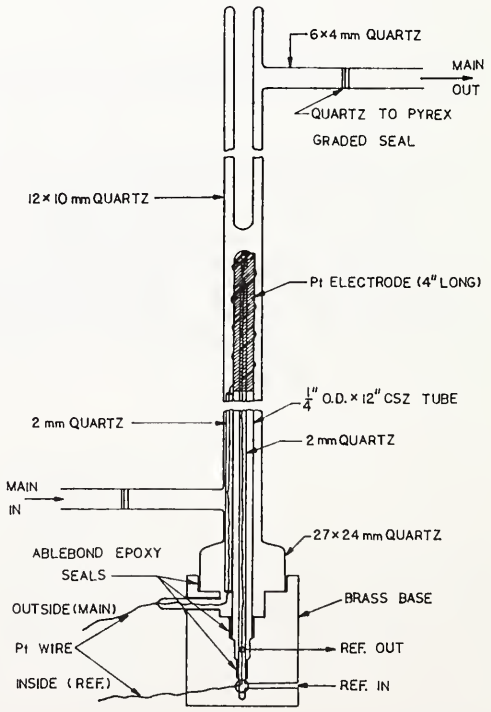


FIGURE 2. *CSZ Pump Design*

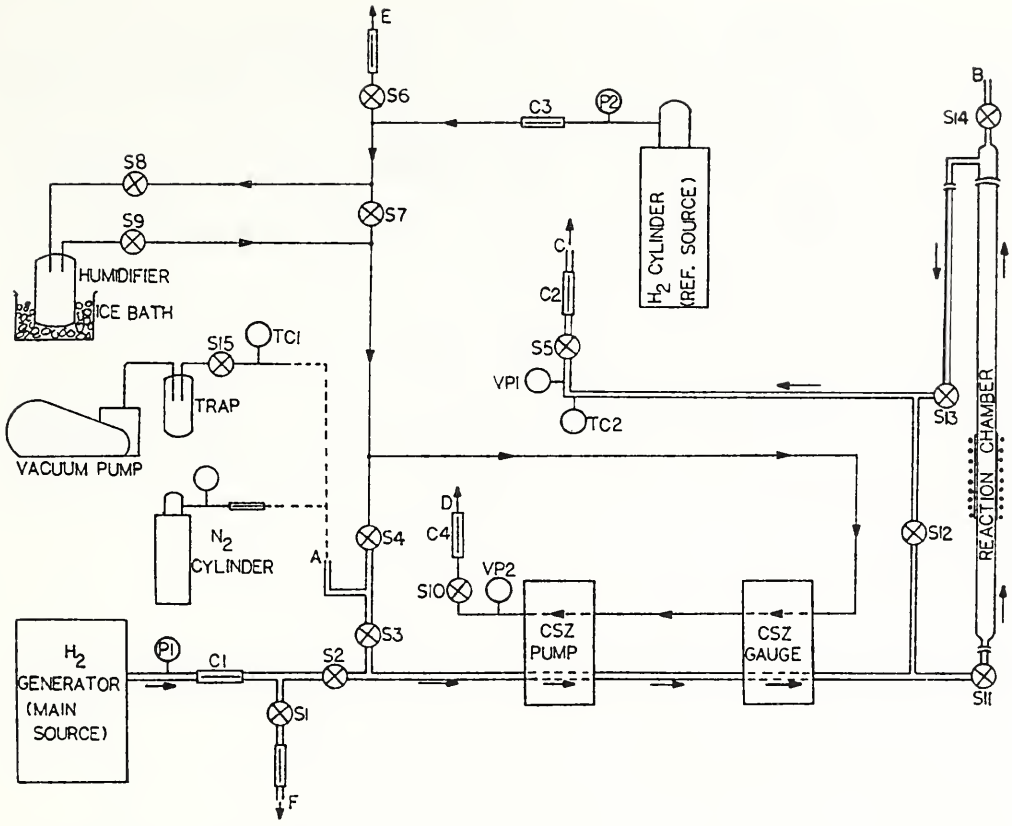


FIGURE 3. *Gas Flow System*

Results and Discussion

3.1 *Control and Measurement of $H_2O(g)$ in $H_2(g)$ Flow*

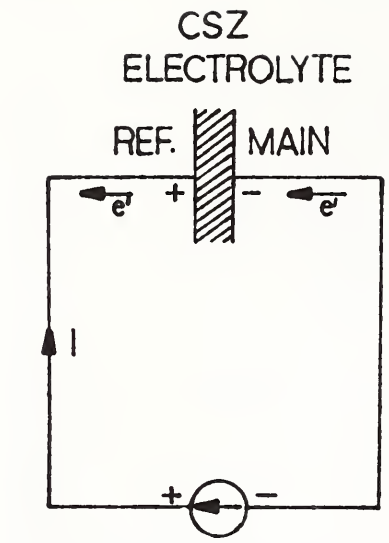
In the control and measurement of H_2O concentrations in an H_2 gas stream, an ultra high purity (UHP) H_2 gas was generated by an Elhygen Mark V hydrogen generator with a built-in palladium diffuser. A cylinder of UHP H_2 was employed as a source of the reference gas. The oxygen activity of the reference gas was established by passing $H_2(g)$ through a water saturator which was kept at 0°C [3] where P_{H_2O} was known to be 6.025×10^{-3} atm [4].

The The H_2O concentration in the main H_2 gas may be altered by applying an electrical potential across the CSZ pump surfaces. In practice, however, a constant current source (Keithley model 227) was used because it provided a very steady and more convenient means of controlling the amount of $H_2O(g)$ to be added to or removed from the gas stream. When a current source is connected to a CSZ pump so that the positive and negative terminals are connected to the reference and main electrodes on the pump surfaces respectively (Figure 4), $H_2O(g)$ is removed from the main gas as follows:

main electrode $$H_2O + V_O^{\cdot\cdot} = 2e' = O_O^X + H_2 \quad (3)$$

ref. electrode $$O_O^X + H_2 = H_2O + V_O^{\cdot\cdot} + 2e' \quad (4)$$

At the main electrode, oxygen removed from H_2O molecules combines with oxygen ion vacancies on the electrode surface and electrons to become electri-



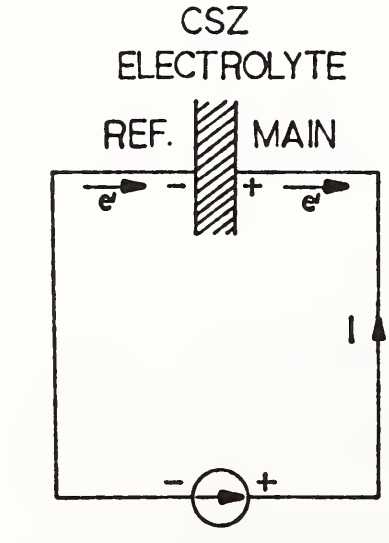
MAIN SIDE:

$$H_2O + V_O^{\cdot\cdot} + 2e' \longrightarrow O_O^x + H_2$$

REF. SIDE:

$$O_O^x + H_2 \longrightarrow H_2O + V_O^{\cdot\cdot} + 2e'$$

(a) REMOVAL OF H_2O



MAIN SIDE:

$$O_O^x + H_2 \longrightarrow H_2O + V_O^{\cdot\cdot} + 2e'$$

REF. SIDE:

$$H_2O + V_O^{\cdot\cdot} + 2e' \longrightarrow O_O^x + H_2$$

(b) ADDITION OF H_2O

FIGURE 4. *Electrode Surface Reactions*

cally neutral oxygen sites. At the reference electrode, oxygen atoms from oxygen sites near the electrode surface react with H_2O and form $H_2O(g)$. This in turn creates oxygen ion vacancies and electrons are released. These electrons flow back to the current source while oxygen ion vacancies migrate to the main electrode surface to provide a continuous supply of $V_O^{\cdot\cdot}$ in reaction (3). To add $H_2O(g)$ to the main gas stream, the polarity between the current source and the CSZ pump electrodes are reversed. The amount of current required to add or remove lppm (10^{-6} atm) of H_2O to or from the main gas stream is 0.1436 Q μA [2] where Q is the flowrate in scc-atm/min.

At a flowrate of 100 cc/min, the H_2O concentration in the main gas stream without pumping (i.e. background level) was found to be ~ 3.3 ppm. About half of the H_2O measured was furnished by the hydrogen supply and was fixed regardless of a flowrate. The remaining H_2O was believed to be the result of outgassing from various glass epoxied and O-ring joints, and it varied inversely with flowrates. When electric current was applied to the pump (Table 1), the H_2O concentrations were shown to be adjustable from 0.0223 ppm to above 100 ppm. The lower and upper limits can be extended further by increasing and decreasing the flowrate respectively. Note that the current required to add 1 ppm of H_2O was 13.29 mA since the flowrate of 100 cc/min at room temperature (22°C) implied that the standard flowrate was (100 × 273)/(273 + 22) = 92.54 scc/min. Figures 5-7 show the plots of measured ppm versus background ± pumped ppm. The pump efficiency decreased significantly after a few ppm of H_2O had been removed from the main gas since there was a near complete depletion of H_2O in the gas stream. When the current was applied in the direction so that H_2O was added to the main

TABLE 1

	Pump Current (μ A)	Pumped ppm	Background ± Pumped ppm	Measured Emf (mV)	Measured ppm
Increase H_2O(g)	1328.63	100	103.31	134.7	105.56
	930.04	70	73.31	144.7	78.18
	664.32	50	53.51	155.7	56.18
	389.59	30	33.31	172.3	33.73
	265.73	20	23.31	184.6	23.59
	132.86	10	13.31	202.9	13.62
	93.00	7	10.31	211.7	10.42
	66.43	5	8.31	219.1	8.37
	53.15	4	7.31	223.6	7.30
	39.86	3	6.31	228.7	6.27
	26.57	2	5.31	234.5	5.27
	13.29	1	4.31	241.7	4.25
	0	0	3.31	250.0	3.31
Remove H_2O(g)	10	-0.753	2.56	262.3	2.28
	13.3*	-1.000	2.31	271.0	1.76
	20	-1.505	1.81	273.5	1.64
	26.57*	-2.000	1.31	297.5	0.795
	30	-2.258	1.05	283.9	1.195
	40	-3.011	0.299	292.0	0.924
	39.86*	-3.000	0.31	347.5	0.177
	50	-3.763	-0.45	393.0	0.045
	60	-4.516	-1.21	396.3	0.041
	70	-5.289	-1.98	406.5	0.0301
	80	-6.021	-2.71	410.5	0.0267
	90	-6.774	-3.46	411.5	0.0259
	100	-7.527	-4.22	415	0.02335
	110	-8.279	-4.97	415.8	0.0228
	120	-9.032	-5.72	416.0	0.0225
	130	-9.785	-6.48	416.4	0.02234
	150	-11.29	-7.98	416.5	0.02233

Q(T = 22°C, P = 1 atm) = 100 cc/min = 92.54 SCC-atm/min

*Extension of data obtained at the same time as data for the addition of H_2O to the background level.

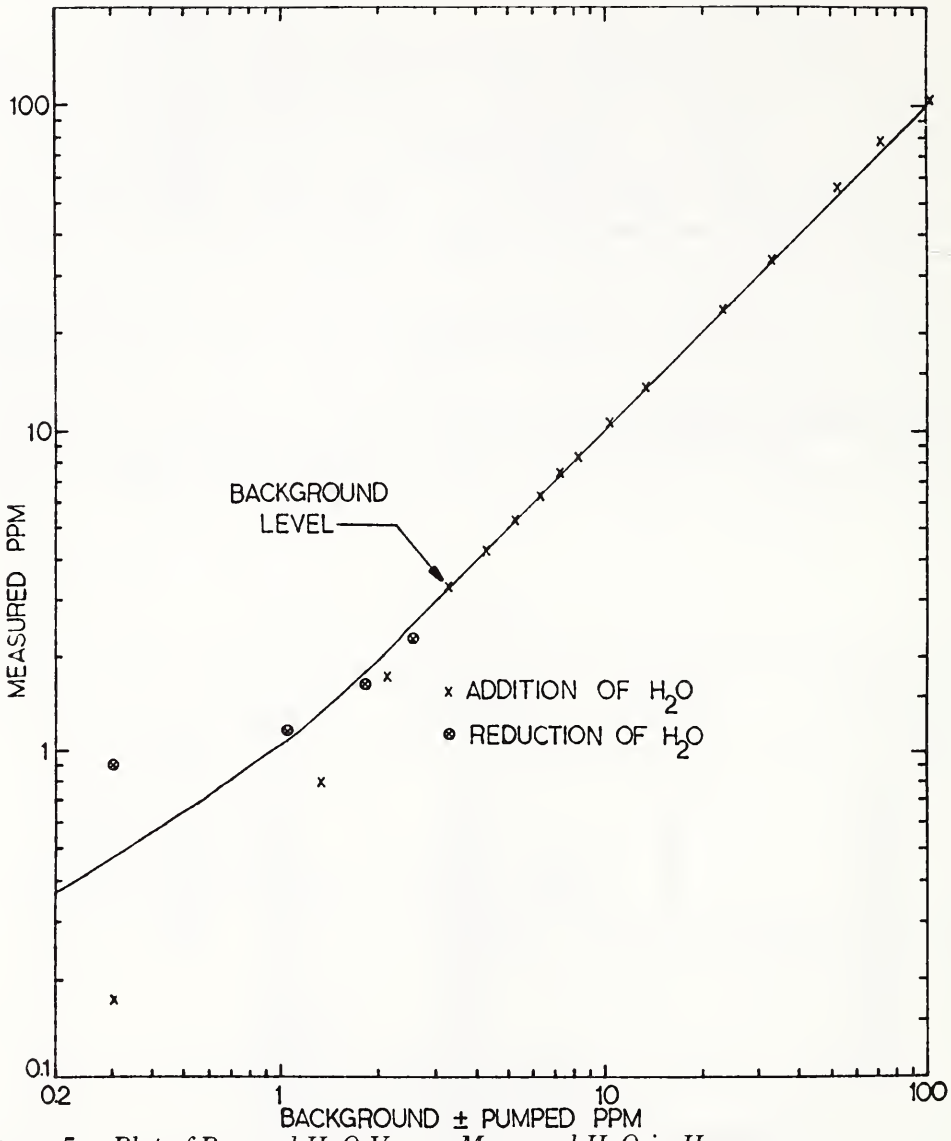


FIGURE 5. *Plot of Pumped H_2O Versus Measured H_2O in H_2*

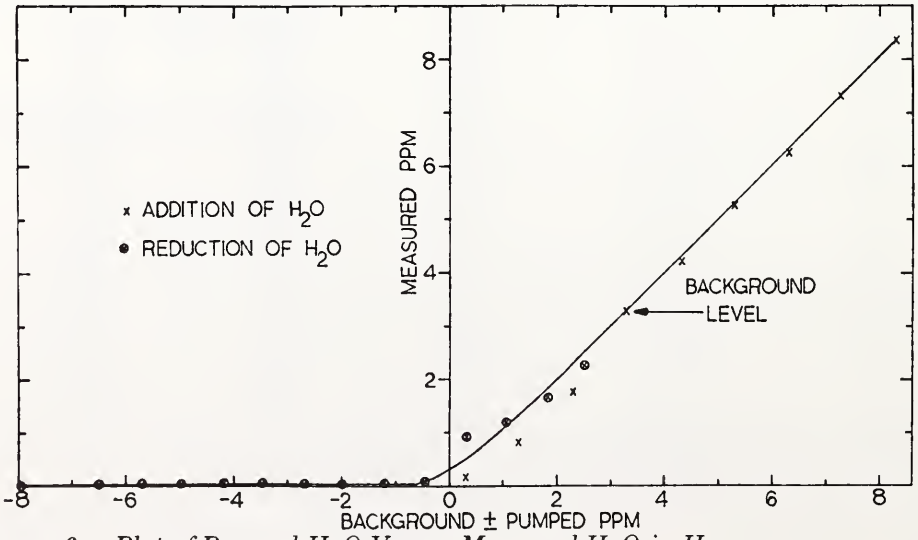


FIGURE 6. *Plot of Pumped H_2O Versus Measured H_2O in H_2*

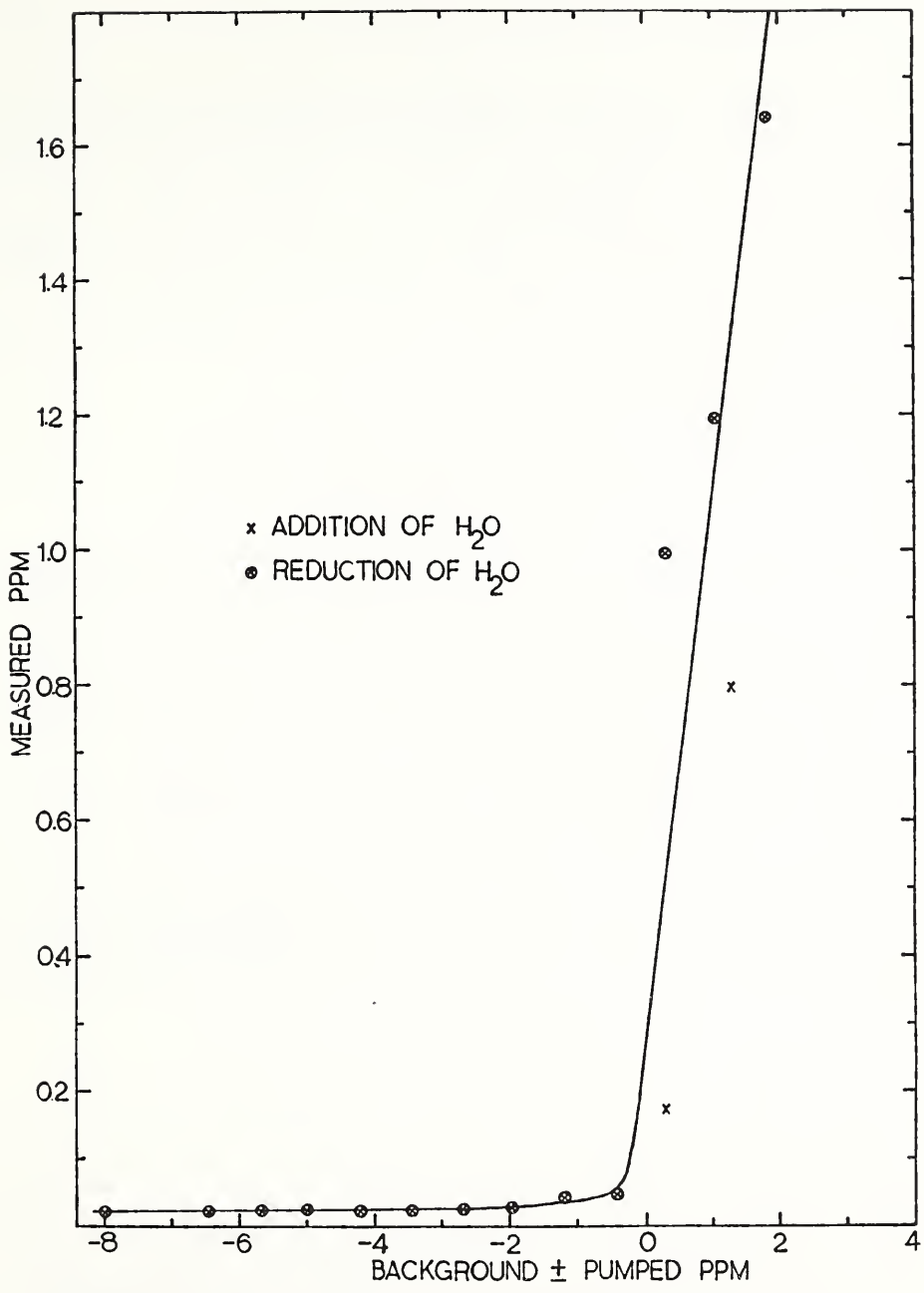


FIGURE 7. *Plot of Pumped H_2O Versus Measured H_2O in H_2*

gas stream, the pump exhibited an almost linear relationship between the pump current and the measured amount of H_2O being pumped. Some discrepancies were observed between 1 and 2.5 ppm since this was the area most sensitive to environmental changes. Fluctuation of H_2 flowrates from the H_2 generator due to line voltage fluctuations, slight variation from 0°C of the ice bath (if the bath temperature was -1°C, for instance, the H_2O level from the reading would be a few tenths ppm higher then the actual level), fluctuation of room temperature, and inaccuracies of the equipment used (voltmeters and ammeter) were all thought to be responsible for the recorded errors.

3.2 *Control and Measurement of O_2(g) in N_2(g) Flow*

The experimental set-up for the control and measurement of oxygen content in N_2 gas stream was essentially the same as that for the control and measurement of H_2O concentrations in H_2 gas. N_2 gas from a UHP-N_2 cylinder was fed into the main path through a capillary tube at point A (Figure 3). Again H_2 gas from a UHP-H_2 cylinder, saturated with H_2O(g) at 0°C was used as a reference gas. Since we wish to interpret the measured emf's in terms of oxygen partial pressure (P_{O_2}), equation (1) was used here. $P_{O_2}$is the equilibrium oxygen partial pressure of (P_{H_2O}/P_{H_2}) ref and is found to be $7.1364933 \times 10^{-33}$ atm at 500°C [4]. Equation (1) thus becomes

$$P_{O_2} = \exp \{ - \frac{4FE}{RT} + 1n(7.1364933 \times 10^{-33}) \} \quad (5)$$

At the flowrate of about 10 cc/min at 22°C (9.25 scc/min), P_{O_2} in N_2 flow was measured to be 1.36×10^{-30} atm at the gauge electrode surface. This was equivalent to P_{H_2O}/P_{H_2} (in flowing N_2) = 0.126 at 500°C [4]. Upon application of electric current to the CSZ pump in the direction to add oxygen to the main N_2 gas stream, P_{O_2} began to rise (Figure 8). Initially oxygen added to the N_2 gas stream combined with trace H_2(g) impurity to form H_2O(g). The rise of P_{H_2O}/P_{H_2} or its equivalent P_{O_2} was gradual but steady as the pump current increased. At about 330 μA of applied current ($P_{O_2} \cong 2.5 \times 10^{-25}$ atm), a sharp rise of P_{O_2} was observed. This was most likely due to the exhaustion of the available H_2(g) in the flowing N_2 gas. The curve then began to flatten at the onset of addition of O_2 to the main gas stream where P_{O_2} is about mid-10^{-6} atm. P_{O_2} could be pumped to about mid-10^{-4} atm at 1000μA pump current. Plots for flowrates of 20, 30 and 50 cc/min yielded a very similar characteristics and little change in values of P_{O_2} and hence are not shown here.

Conclusion

A CSZ pump and a CSZ gauge were designed, fabricated and built into a gas flow system to control and measure oxygen partial pressures (or H_2O) in N_2 or H_2 gas streams. In streaming H_2, H_2O(g) content could be varied from 0.0223 ppm (equivalent $P_{O_2} = 1.067 \times 10^{-43}$ atm) to over 100 ppm (equivalent $P_{O_2} < 1.97 \times 10^{-36}$ atm) with a background level at 3.3 ppm at 100 cc/min flowrate. This range can be expanded by varying flowrates. In an N_2 flow, the background (equilibrium) oxygen partial pressure was found to be 1.36×10^{-30} atm at 10 cc/min flowrate. When current was applied to the pump to add oxygen to streaming N_2, P_{O_2} rose gradually with pump current until it reached $\sim 2.5 \times 10^{-25}$ atm. A sharp rise of P_{O_2} due to exhaustion of H_2(g) in N_2 was then observed. The curve began to flatten out at $P_{O_2} \cong$ low-mid 10^{-6} atm due to the onset of addition of oxygen to N_2. P_{O_2} of about mid 10^{-4} atm were achieved with 1000 μA pump current. Since the breakpoint from low to high P_{O_2} value when pumping oxygen into N_2 apparently depends on the H_2 concentration in the N_2, one should be able to adjust the breakpoint upward by adding more H_2 to the N_2. Observe that below the breakpoint the curve (Figure 8) is nearly linear such that one order of magnitude change in P_{O_2} corresponds to $\sim$ 50 μA pump current.

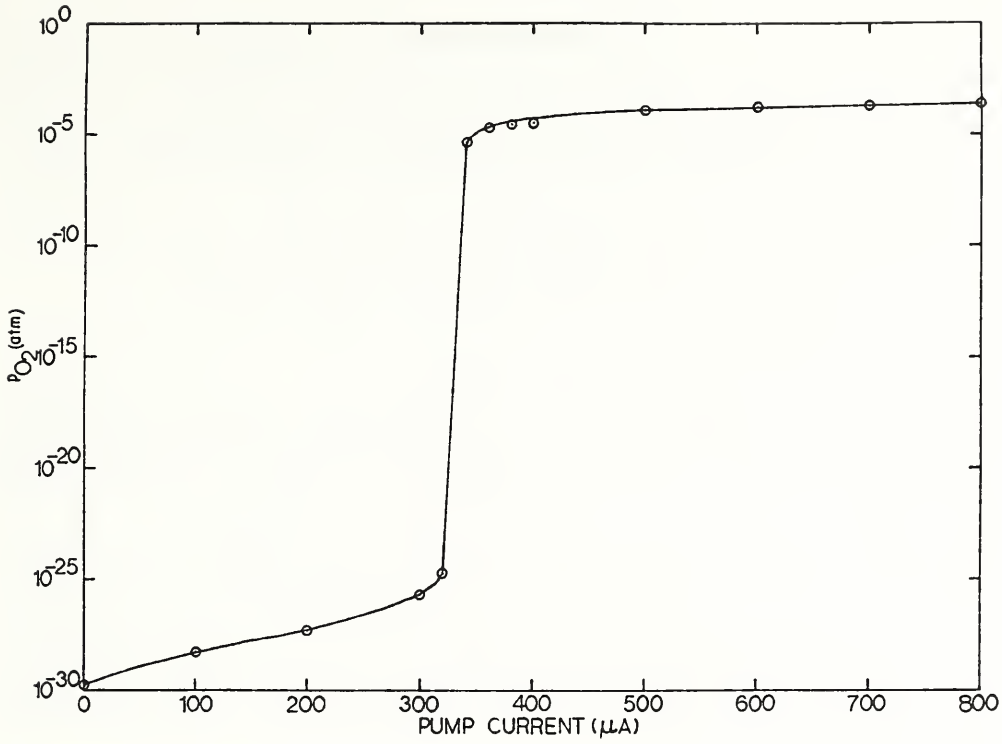


FIGURE 8. *Plot of Pump Current Versus Oxygen Partial Pressure in N_2*

Thus one can vary the oxygen partial pressure in flowing N_2 or H_2 to create a wide range of conditions for oxidation or reduction in a reactor located as shown in Figure 3.

Acknowledgment

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ENTOMOLOGY

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ABSTRACTS

The Influence of Nutrient Levels on the Colonization of Tires by Mosquitoes. William J. Berry, Vector Biology Laboratory, University of Notre Dame, Notre Dame, Indiana 46556.—Previously unexploited tires were placed in tireyards and woodlots in shaded and exposed areas. The rate of colonization of the new habitats by tire inhabiting mosquitoes was determined by sampling the tire contents from June through September. The rate of colonization was found to differ greatly between species and also was influenced by the amount of exposure to the sun and the amount of leaf litter present in the tire. *Culex restuans* was found to be the first species to exploit the new habitat; *Aedes atropalpus* and *Aedes triseriatus* were slower to colonize and occurred in fewer numbers than *Culex restuans.*

Electrophoretic Separation of Northern Indiana Culex Species. Bartholomew G. Corsaro and Leonard Munstermann, University of Notre Dame, Notre Dame, Indiana 46556.—The Saint Joseph County (Indiana) Mosquito Abatement Program conducts routine surveillance for nuisance and vector mosquitoes with sixteen New Jersey light traps located in major population centers throughout the county. The primary purpose of the project is to assess the risk of Saint Louis Encephalitis in the county. Unfortunately, *Cx. pipiens*, the major vector of SLE, is virtually impossible to separate from adult *Cx. restuans* because characteristic scales are rubbed off either by age or the trapping device.

Starch gel electrophoresis was examined as a method to separate these species. Electrophoresis was performed on adult specimens of *Cx. pipiens*, *restuans*, *territans*, and *salinarius*, that were collected and identified as larve.

Twenty-seven enzymatic assays were attempted, of these, seventeen produced repeatable banding patterns. Six loci (IDH, GPI, ME, AK, ACO, ODH) separated *Cx. restuans* from *Cx. pipiens* and *salinarius*; while only one loci (ME) separated *Cx. salinarius* from *Cx. pipiens*. No enzymes separated *Cx. territans* though this is umimportant since it posesses reliable morphological characters. Most importantly, *Cx. pipiens* could reliably be separated from *Cx. restuans.*

This information was used to determine the proportion of each *Cx.* species in five of the sixteen light traps in the surveillance program. A CDC net bag was attached to the New Jersey traps once a week from June through August 1982. The Culex mosquitoes collected were identified by starch gel electrophoresis using the enzyme systems mentioned above. These techniques allowed us to determine the density of *Cx. pipiens* in relation to *Cx. restuans*, and the time of first appearance of *Cx. pipiens* in the light traps.

On the Genetic Basis of Non-reciprocal Fertility among Members of the Aedes (Stegomyia) scutellaris Group (Diptera, Culicidae). Vas Dev and Karamjit S. Rai,

Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.—*Aedes* (*Stegomyia*) *scutellaris* group is composed of over 30 species. Of these, *Aedes malayensis* and *A. alcasidi* are very closely related species separable mainly by the distinct claspette of the male genitalia. Interspecific crossing experiments revealed that there existed only a unidirectional compatability between these two species. 100% of *A. malayensis* females were inseminated by males of *A. alcasidi* and gave viable progeny. However, in the reciprocal cross, 100% of *A. alcasidi* females were inseminated but all eggs laid were sterile.

Hybrid progeny obtained from the cross using *A. malayensis* females and *A. alcasidi* males were backcrossed to either parent. *A. malayensis* females inseminated by hybrid males laid viable eggs. However, eggs laid by the *A. alcasidi* females inseminated by the hybrid males, were all sterile. The hybrid females laid viable eggs irrespective of the type of species males use. Thus, it is evident that it is always the *A. alcasidi* females which lay sterile eggs in the interspecific crosses.

Of several possibilites, the chromosomal basis of this unidirectional compatibility appears unlikely for the following reasons. *A. alcasidi* females when inseminated by hybrid males should be able to recognize its homologous chromosomes (barring crossing over) resulting in some degree of fertility depending on the number of chromosomes involved. Besides, a proportion of females, recovered from the backcross (hybrid females × *A. alcasidi* males), when further backcrossed to *A. malayensis* males, are expected to lay sterile eggs but all laid viable eggs based on the 35 families raised. From this crossing scheme, it appears that the genetic basis of unidirectional fertility is of cytoplasmic origin and independent of nuclear chromosomes.

Ice Nucleating Proteins from the Freeze Tolerant Queens of the White Faced Hornet, Vespula maculata. JOHN G. DUMAN, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.—In late summer 100-200 new queens are produced in the nest of the white faced hornet, *Vespula maculata*. After being inseminated these future queens overwinter individually in logs, etc. During this overwintering period they are freeze tolerant (able to survive the freezing of their extracellular fluids). In spite of the presence of high levels of the cryoprotectant glycerol, the supercooling points of the overwintering queens are quite high (about -5°C) because of the production (beginning in the pupal stage) of ice nucleating proteins. These hemolymph proteins function to inhibit supercooling. This insures that ice formation occurs at fairly high temperatures and that it is initiated in the hemolymph. This is important because freezing after extensive supercooling can lead to intracellular ice formation which is lethal, even for freeze tolerant insects.

An ice nucleating protein was purified from the hemolymph of pupal *V. maculata* queens using ion exchange (DEAE-Sephadex), gel filtration (LKB-Ultogel-ACA-54) and preparative polyacrylamide gel electrophoresis. This is the first ice nucleating protein to be purified. The protein has a molecular weight of 74,000, as determined by SDS-PAGE. The amino acid composition of the protein is interesting because it contains ~20 mol % glutamic acid and/or glutamine. This along with the other hydrophilic amino acids (Asp, Lys, Asn, Arg, Ser, Thr) makes the protein quite hydrophilic. Hydrophilic amino acids account for 54% of the amino acids present in the protein. It is likely that the hydrophilic nature of the protein allows it to order water in a special way so that small clusters of water molecules (embryo crystals) are aggregated around the protein. Once these embryo crystals reach a critical size they then seed the solution.

Hormonal Regulation of Antifreeze Protein Production in Larvae of the Beetle, Dendroides canadensis. KATHLEEN L. HORWATH and JOHN G. DUMAN, Department of Biology, University of Notre Dame, Notre Dame, Indiana 46556.—Larvae of the beetle *Dendroides canadensis* accumulate protein antifreezes during the winter which depress their hemolymph freezing and supercooling points, thereby enhancing their overwintering survival. This study describes experiments investigating the endocrine control of these antifreeze proteins. *D. canadensis* which were collected in the early fall, prior to the initiation of cold hardening processes, were treated with either 3.3 or 6.6 μg juvenile hormone I topically in acetone and maintained for 21 days under normally non-inductive acclimation conditions (16 light/8 dark, 20°C). Hormone treated animals significantly elevated the levels of antifreeze protein in their hemolymph compared to those of acetone treated and untreated controls or animals measured on the day of collection.

D. canadensis treated with the anti-JH compound precocene II (P2) in acetone for 24 hr at a concentration of 20 $\mu g/cm^2$ (a dose below the LD_{50} for 'behavioral' survival) and then maintained under acclimation conditions conducive to antifreeze protein production (8 light/16 dark, 20°C) for 2 weeks failed to elevate levels of antifreeze. Acetone treated control animals accumulated a significant concentration of antifreeze protein. *D. canadensis* were also treated with 20 and 150 $\mu g/cm^2$ P2 (a dose below the LD_{50} for 'gross' survival) followed by acclimation to short (8 hr) photoperiod at 10°C. All animals receiving the higher P2 dosage failed to elevate antifreezes while only 42.9% of the individuals treated with the lower dosage initiated antifreeze protein production. In contrast, over 80% of untreated and 70% of acetone treated controls responded to these inductive acclimation conditions by elevating antifreeze concentrations.

These results indicate that juvenile hormone participates in the seasonal control of antifreeze protein production in *Dendroides canadensis*. Since this species does not enter a diapause state prior to or throughout the winter this is the first evidence establishing a direct hormonal mechanism involved with insect cold hardiness.

This research has been supported in part by a grant from the Indiana Academy of Science to KLH.

Evaluation of a Method of Estimating Yields of Alfalfa under Insect Stress. MARK A. ZAJAC and M. CURTIS WILSON, Department of Entomology, Purdue University, West Lafayette, Indiana 47907.——Investigators have suggested that alfalfa yields may be accurately estimated to within 10% of actual yields using plant height as the sole estimating variable. They developed a fourth order polynomial equation relating crop height to yield. This equation was assumed to be accurate in full stands of alfalfa (35 or more stems per square foot.).

Data from research conducted over the past 10 years in Indiana on the effects of the potato leafhopper, *Empoasca fabae* (Harris), on alfalfa yields show a high correlation between insect populations and crop height. Consequently, yields estimated through the formula and potato leafhopper populations were also highly correlated. However, insect populations showed a low correlation with harvested yields. Crop height and harvested yields also showed little correlation. Data from 11 field studies support the conclusion that crop height is of little value in estimating yields of alfalfa under insect stress.

Ecology of Vectors of Jamestown Canyon Virus: Seasonal Succession of Hematophagous Diptera at Kingsbury State Fish and Wildlife Area, Laporte County, Indiana 1982

Robert D. Boromisa and Paul R. Grimstad
Department of Biology
University of Notre Dame
Notre Dame, Indiana 46556

Introduction

This paper is the initial report of arthropod collections made as a part of a long-term investigation into the ecology of vectors of Jamestown Canyon virus (JCV) in the upper Midwest. A primary goal is to identify the arthropod vector(s) of JCV in the natural disease cycle.

Jamestown Canyon is a serotype of Melao virus (Family Bunyaviridae) of the California Serogroup, and has recently been shown to cause viral encephalitis in humans (4). White-tailed deer are considered to be the primary host of the virus (6). Despite the fact that virus isolations have been made from many areas of the United States (4), little is really known about the natural disease cycle of JCV. Before any vector can be identified, we need to know which arthropods are present at the study site, in what numbers, and at what time of the season they are present. An earlier serosurvey of Indiana residents (5) had identified a number of JCV foci of infection in northern Indiana, especially in the Kingsbury area. We anticipated that collections from that region would furnish hematophagous arthropods for virus isolation. The collection of bloodfed specimens would also enable us to identify bloodmeal sources and establish host-vector relationships. This report will describe the seasonal succession of numerous species of hematophagous Diptera collected at Kingsbury in the summer of 1982.

Materials and Methods

Study site

Kingsbury State Fish and Wildlife Area is an Indiana Department of Natural Resources (IDNR) regulated site that has extensive areas of oak woods, open prairies, marshlands, and a tamarack lake. This site is ecologically and geologically characteristic of the Northwestern Prairie and Wetlands Division, one of eight natural divisions described for Indiana (8). Kingsbury is the IDNR area in closest proximity to the most intense focus of human infection by JCV in Indiana, based on an earlier serosurvey (5). An earlier serosurvey of deer from Kingsbury detected numerous animals with neutralizing antibody to JCV (1). Thus, Kingsbury appears to be an ideal site for this study.

Collection sites

Collection sites were chosen for their ecological differences and their spatial distribution within the study site. The six collection sites were: 1) along the Kankakee River in a low lying wooded area on the southern edge of Kingsbury that floods in the spring and periodically during the summer, 2) in the periphery of a heavily wooded oak forest in the interior of Kingsbury, 3) by the tamarack lake in the eastern reaches of the study site, 4) near a large drainage canal in a shrubby, open area in the central Kingsbury site, 5) in a damp, low lying area near an oak-cherry forest, and 6) by a permanent marshy area near small oak and pine woodlots in an open prairie near the headquarters.

Collection methods

Hematophagous Diptera were collected using seven different methods: 1) CO_2-baited miniture light traps,[1] 2) a lightweight battery powered aspirator (10), 3) a CO_2-baited cone trap (2), 4) resting boxes (3), 5) oviposition traps (7), 6) a Malaise trap,[1] and 7) human-baited mosquito biting collections. The lightweight battery powered aspirator was not used at the tamarack lake collection site since the ground cover was inadequate to shelter resting mosquitoes. Human-baited mosquito biting collections were not conducted at the tamarack lake because of the frequently windy conditions that made mosquito flight impossible. The CO_2-baited cone trap and Malaise trap were used only at site 2. These traps are of a permanent nature and could not be moved easily.

All seven collection methods were used at the other collection sites. Light trap collections were made once a week from the last week in May until the end of the collecting season in mid-September. The CO_2-baited cone trap and aspiration collections were initiated in early June. Mosquitoes were aspirated from shrubby ground cover in the immediate vicinity of the light traps. Resting boxes and oviposition traps were set up in mid-July. The Malaise trap was put into operation beginning in early August. Human-baited mosquito biting collections did not begin until mid-August. Biting mosquitoes were captured in 8-dram vials, returned to the laboratory and identified (12) to species.

Specimen handling

All specimens were brought back to the laboratory and the non-hematophagous arthropods were sorted out and discarded. Live tabanid specimens from each day's collection were grouped together and frozen at -70°C until identified at a later time. The tabanids were identified (11) on ice prior to storage to preserve potential virus activity and refrozen at -70°C for future virus isolation. Dead specimens were identified and used for voucher specimens. Live mosquitoes were killed by freezing, held on ice for identification (12), then pooled in groups of 50 or fewer specimens. Mosquito pools were based on collection day and method of collection; these pools were also frozen at -70°C for future virus isolation. Dead mosquitoes were identified (12) and used as voucher specimens. *Simulum vittatum* and *Simulum tuberosum* larvae and pupae were collected from the central drainage canal in Kingsbury in early September. Specimens were placed in 70% ethanol and identified (9) in the laboratory. Simuliid specimens were not pooled and frozen for virus isolation since so few specimens were collected.

Results

The 15,340 specimens collected at the 6 sites during the spring and summer of 1982 included 11 genera representing 38 species of Diptera belonging to three families.

Light traps

The five species of mosquitoes listed in Table 1 represented 98% of the specimens collected with the CO_2-baited light traps. *Aedes vexans* and *Aedes trivittatus* were the most numerous mosquito species collected followed by *Coquillettidia perturbans* and the *Culex pipiens/restuans* complex. *Aedes vexans* was the most numerous species in light trap collections from late May through mid-July.

[1]Bioquip Products, Santa Monica, California 90406, U.S.A.

TABLE 1. *Mosquito light trap collections at Kingsbury State Fish and Wildlife Area in 1982.*

Species Collected	May	June				July				August				September		TOTAL
	4*	1	2	3	4	1	2	3	4	1	2	3	4	1	2	
Aedes vexans	12	448	99	283	84	419	1155	212	340	97	514	567	194	106	38	4155
trivittatus	3	163	83	253	188	382	983	540	673	531	415	189	251	186	96	4936
stimulans	4	22		1	4	9	4									44
Coquillettidia perturbans		50	27	44	18	169	133	16	51	34	29	113	121	134	24	963
Culex pipiens/restuans	2	4					29	13	30	50	218	111	31	32	15	535
TOTAL	26	687	209	581	294	979	2304	781	1094	712	1176	567	597	458	173	10633

*week of the month

TABLE 2. *Mosquito aspiration collections at Kingsbury State Fish and Wildlife Area in 1982.*

Species Collected	June			July				August				September		TOTAL
	1*	2	3	1	2	3	4	1	2	3	4	1	2	
Aedes vexans	20	107	86	150	68	556	254	185	309	114	70	27	19	1965
trivittatus		64	35	40	15	154	63	52	26	19	8	7	2	485
stimulans	61	9	3	12	4	6	4							99
Culex territans	13	8	11	17	14	30	17	53	21	45	15	18	8	279
pipiens/restuans	9	6	4	5		12	5	1	62	14	1	3	4	123
Uranotaenia sapphirina							4		8	13	33	26	28	113
Coquillettidia perturbans	7	9	4	6	1	4	2	3	7	5	6	3	2	59
TOTAL	110	203	143	230	102	762	349	294	433	210	133	84	60	3123

*week of the month

Aedes trivittatus was the most numerous species collected from late July until the end of the collecting season. *Aedes stimulans* persisted through mid-July. *Coquillettidia perturbans* were present throughout the season with peak numbers obtained in early July and late August. *Culex pipiens/restuans* were collected in light traps early in the season, disappeared, and then reappeared in mid-July and peaked in mid-August.

Other mosquito species collected were: *Aedes abserratus*, *Aedes canadensis*, *Anopheles punctipennis*, *Anopheles quadrimaculatus*, *Anopheles walkeri*, and *Uranotaenia sapphirina*. There were no bloodfed mosquitoes collected in light traps.

Aspiration collections

The seven species of mosquitoes listed in Table 2 represented 95% of all specimens collected in this manner. The numbers of males and females of each species were combined to give the totals listed in Table 2. Of the specimens collected, 30% were male and there were 560 bloodfed mosquitoes collected. *Aedes vexans* was the most numerous species collected except in the early and late parts of the collecting season. There were relatively few *Ae. trivittatus* collected compared to the numbers of *Ae. vexans*. The total number of *Ae.* vexans was lower than that obtained in the light traps; numbers of *Ae. trivittatus* and *Coq. perturbans* were greatly reduced from light trap collections. However, this was the only method of collecting *Culex territans* and *Uranotaenia sapphirina*, in numbers. Other mosquito species collected were: *Aedes abserratus*, *Aedes canadensis*, *Aedes cinereus*, *Anopheles punctipennis*, *Anopheles quadrimaculatus*, and *Culiseta inornata*.

Co_2-baited cone trap

The CO_2-baited cone trap collected large numbers of tabanid species but collected only a few mosquitoes and no other hematophagous Diptera. Tabanids in the genus *Tabanus* were the most numerous followed by *Hybomitra* and *Chrysops*. The 18 species of Tabanidae collected are listed in Table 3. The tabanids collected were seasonal in distribution. Only *Tabanus lineola* and *Tabanus similis* were collected throughout the summer. However, there were relatively few tabanids collected in the month of July because the original trap site was rototilled by IDNR personnel, thereby, destroying all foilage in the immediate area. The trap was moved to several other locations within the collection site until good collections were obtained.

Resting boxes

There were only 50 mosquitoes collected in six resting boxes. The four species of mosquitoes collected in this manner are listed in Table 4. This method was best for collecting *Anopheles quadrimaculatus*.

Ovipositions traps

A total of 40 oviposition traps were set up at all collection sites. Only 12 of the oviposition trap paddles containing eggs were collected. The remainder of the oviposition paddles were lost from the oviposition traps due to unknown natural causes. Presence of eggs on the balsawood paddles indicated the presence of *Aedes triseriatus* complex members, either *Aedes triseriatus*, *Aedes hendersoni*, or both.

Malaise trap

The Malaise trap was not effective in catching hematophagous Diptera. Future

TABLE 3. *Tabanid collections at Kingsbury State Fish and Wildlife Area in 1982.*

Species collected	Month Collected				Total
	June	July	August	September*	
Hybomitra					
lasiophthalma	88	1			89
epistates	19	3			22
spp.			1		1
Tabanus					
vivax		1	125		129
lineloa	14	19	31	1	65
similis	24	4	7	6	41
pumilus	10				10
quinquevittatus			7	2	9
trimaculatus	3	1			4
atratus	3				3
marginalis		1	1		2
spp.			25		25
Chrysops					
cincticornis	3	1			4
univittatus		1	3		4
niger	2				2
indus	1				1
callidus	1				1
cuclux	1				1
frigidus	1				1
vittatus			1		1
TOTAL	169	31	204	12	416

*two weeks only

modifications to the trap will include CO_2-baiting and or changing the color of the trap from its present white color to varying shades of green to make the trap less noticeable to the flying insects.

Human-bait collections

The results of the human-bait collections are listed in Table 5. A total of 200 mosquitoes were collected at four different collection sites on four different days.

TABLE 4. *Resting box collections at Kingsbury State Fish and Wildlife Area in 1982.*

Species Collected	Per Cent Collected		
	male	female	TOTAL
Anopheles quadrimaculatus	52	12	64
Aedes trivittatus	24		24
Aedes vexans	8		8
Anopheles punctipennis	2	2	4

TABLE 5. *Human-bait collections at Kingsbury State Fish and Wildlife Area in 1982.*

Species Collected	Per Cent of Total
Aedes vexans	49
Aedes trivittatus	39
Aedes triseriatus	6
Coquillettidia perturbans	4
Anopheles quadrimaculatus	2

Discussion

Collectons that are representative of the field populations are needed for the accurate determination of the seasonal succession of hematophagous Diptera. The different collection methods used in this study clearly demonstrated that not all collection methods yielded the same results and that species numbers varied considerably among the trapping techniques. Thus, a variety of collection methods are essential to minimize collection bias and provide representative collections.

A seasonal succession of hematophagous Diptera was noted in the spring and summer of 1982. No single species predominated at any one trap site all season. The absence of early spring (snow melt) *Aedes* species is striking in our collections. Unfortunately, the early rapid snow-melt was followed by a cool dry period that resulted in vernal pools drying before adult emergence. We were not sure of the effect this might have had on JCV transmission at Kingsbury in the summer of 1982, since earlier studies in Wisconsin (6) indicated JCV was transmitted in the late spring to white-tailed deer. However, our analyses of deer bloods collected in October and November of 1982, indicated approximately 60% of the deer that were now two summers old, had antibodies to JCV. A 1982 serosurvey indicated that 4% of yearling deer had antibody. Thus, it appears that approximately 56% of the deer that spent their second summer in the northwestern area of Indiana and were harvested this fall at Kingsbury, seroconverted to JCV. Virus isolation studies and analyses of bloodmeals are in progress to identify potential vector species and establish host-vector relationships.

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A Mosquito Larval Survey of Vanderburgh County, Indiana

VIVIE E. DUNN
Public Health Entomologist
Vector Control, Division of Sanitary Engineering
Indiana State Board of Health
Indianapolis, Indiana 46206
and
VAN G. GHORMLEY
Supervisor Vector Control Section
Evansille-Vanderburgh County Health Department
Evansville, Indiana 47708

Introduction

Evansville, with a population of 130,496 (1980 census), is the largest city in southern Indiana. Historically, it has been the site of outbreaks of the mosquito borne disease, St. Louis Encephalitis (SLE). *Culex pipiens* complex is considered to be the main vector of SLE in southwestern Indiana (Newhouse and Siverly, 1966). Although efforts had been directed towards mosquito control in Evansville for several years, an attempt to base a mosquito control program primarily on larvaciding *Culex* breeding sites, was begun in 1974, by the Evansville-Vanderburgh County Health Department. Much information on the mosquitoes of Vanderburgh County was gathered, during the effort to identify *Culex* breeding sites. Only 15 species of mosquito were reported from Vanderburgh County in a list of county records in *Mosquitoes of Indiana* (Siverly, 1972). The objectives of this paper are to update Siverly's list and to summarize the 1974-1979 larval surveys.

Materials and Methods

Vanderburgh County, located in southwestern Indiana, occupies 241 square miles, within the physiographic subdivision known as the Wabash lowland (Kelly, 1976). Much of the terrain is on uplands, with gently sloping to steep soils. Elevation varies from 357 feet above sea level at the Ohio River near the mouth of Bayou Creek, to 600 feet above sea level in the west central part of the county. The bottom lands, including those along the Ohio River, are subject to flooding. The land that is nearly level in the county, is divided between urban uses and grain farming. Most of the larval collections were made in the Greater-Evansville Area. Evansville, with a population of 168,772 (1970 census) sits in the southern part of the county, along the Ohio River.

Larval samples were taken with a standard 460 ml. white dipper and transported to the Evansville-Vanderburgh County Health Department for species determination. Several identifications were made from adults reared from immature stages. Table 1 indicates the period of time when most collections were made during the six seasons, 1974 through 1979. A few collections were made in April in the 1977 through 1979 seasons.

Most breeding sites were located when a mosquito complaint was investigated, during the earlier seasons in this study. Habitats were described in order to identify *Culex* breeding sites for surveillance and control.

Results and Discussion

A total of 1,117 samples was collected as recorded in Table 1. A total of

TABLE 1. *Period of time when larval samples were collected, number of larval samples collected, and number of individual larvae identified in Vanderburgh County, IN, during 1974 through 1979 larval surveys.*

Period of time, when samples collected.			No. of samples collected.	No. of larvae identified.
Year:	From:	To:		
1974	July 10	September 5	37	1677
1975	July 1	October 3	311	6535
1976	May 24	October 1	195	8010
1977	June 2	October 7	276	15875
1978	June 2	September 28	225	8645
1979	June 29	August 24	73	2438
		Totals:	1117	43180

TABLE 2. *Species of mosquitoes collected in Vanderburgh County, IN, during 1974 through 1979 larval surveys.*

	Year of collection						
Species	+ + 1972	1974	1975	1976	1977	1978	1979
Aedes							
aegypti	–	–	X+	–	X	–	–
canadensis	X	–	–	–	–	–	X
cinereus	–	–	–	–	–	–	X+
dupreei	–	–	–	–	–	–	X+
sollicitans	X	–	X	–	–	X	–
sticticus	X	–	X	–	–	–	–
stimulans	–	–	X+	–	–	–	–
triseriatus	–	–	–	–	X+	X	X
trivittatus	X	–	X	–	X	–	–
vexans	X	X	X	X	X	X	X
Anopheles							
punctipennis	X	X	X	X	X	X	X
quadrimaculatus	X	X	X	X	X	X	X
Culex							
erraticus	X	–	X	–	–	–	–
pipiens complex	X	X	X	X	X	X	X
restuans	X	X	X	X	X	X	X
salinarius	X	–	X	–	X	X	X
tarsalis	–	X+ + +	X	X	X	X	X
territans	X	X	–	X	X	X	–
Culiseta							
inornata	X	–	–	–	–	X	X
Psorophora							
ciliata	X	–	X	–	X	X	X
columbiae	–	X+	X	X	X	X	X
cyanescens	–	–	–	–	X+	–	X
ferox	–	–	–	X+	X	X	–
howardii	–	–	–	–	X+	–	X
Uranotaenia							
sapphirina	X	X	X	–	–	–	–

\+ New records.

\+ + Reported by Siverly, 1972.

\+ + + One adult *Cx. tarsalis*, collected from daytime resting site in October, 1964, was reported by Newhouse and Siverly, 1966, but was not reported in Siverly's 1972 publication.

43,180 individual larvae was identified during the six mosquito control seasons 1974 through 1979.

A listing of the mosquito species found in Vanderburgh County is presented in Table 2. During the six years, 25 species were identified, including *Aedes aegypti* (Linnaeus), *Aedes cinereus* Meigen, *Aedes dupreei* (Coquillett), *Aedes stimulans* (Walker), *Aedes triseriatus* Say, *Culex tarsalis* (Coquillet), *Psorophora columbiae* (Dyar and Knab), *Psorophora cyanescens* (Coquillett), *Psorophora ferox* (Humboldt), and *Psorophora howardii* Coquillett.

Table 3 lists the habitats of the mosquito species collected during the 1974 through 1979 summers. *Culex pipiens* complex had the broadest habitat range.

TABLE 3. *Habitats of the mosquito species collected in Vanderburgh County, IN, during 1974 through 1979 larval surveys.*

HABITATS	Ditch (fresh)	Ditch (polluted)	Drainage ditch (fresh)	Drainage ditch (polluted)	Roadside ditch (fresh)	Roadside ditch (polluted)	Slough (brackish)	Slough (polluted)	Old canal bed	Woodland pool	Temporary pool (fresh)	Temporary pool (polluted)	Permanent swamp (wooded)	Permanent pool (brackish)	Permanent pool (polluted)	Golf course pool (fresh)	Cattail marsh	Creek (pools)	Lagoon (polluted)	Sewage treatment plant	Sand filter	Tires	Basement	Metal cabinet	Barrel	Footprints	Wheel tracks	Hole in tree stump
SPECIES																												
Aedes																												
aegypti			X																									
canadensis											X																	
cinereus		X																										
dupreei										X																		
sollicitans							X						X															
sticticus			X																									
stimulans	X																											
triseriatus					X																							X
trivittatus					X						X																	
vexans			X	X	X	X			X		X		X				X	X							X	X	X	
Anopheles																												
punctipennis			X	X	X	X		X									X	X										
quadrimaculatus			X	X	X	X					X		X				X			X		X					X	
Culex																											X	
erraticus																X												
pipiens complex			X	X	X	X		X	X		X	X	X		X				X	X	X	X	X	X			X	
restuans			X		X			X			X								X				X				X	
salinarius			X	X	X			X																				
tarsalis					X	X		X			X								X								X	
territans					X	X		X		X	X		X															
Culiseta																												
inornata			X							X																	X	
Psorophora																												
ciliata			X								X														X		X	
columbiae			X	X	X	X					X		X				X									X	X	
cyanescens											X																	
ferox			X								X		X									X						
howardii								X			X																	
Uranotaenia																												
sapphirina			X		X						X																	

Roadside and drainage ditches, and wheel tracks were the three most frequently described habitats in Vanderburgh County during this study.

Table 4 presents larval associations of the species collected. *Aedes vexans* and *Culex pipiens* complex were found in association with 14 other species. *Anopheles punctipennis* and *An. quadrimaculatus* were found in association with 11 other species. *Culex erraticus* was collected with only one other species, *An. quadrimaculatus*, and *Psorophora howardii* was collected with only one other species, *Ae. vexans*.

Here it would be pertinent to make some comments on the rainfall in Evansville during this study. It is interesting to note that June through August of 1977, was above average in rainfall, with six days of over an inch of rain. That makes 1977, the wettest summer during the six years of this study. *Psorophora* eggs are laid on moist soil and then hatch after summer rains. Five species of *Psorophora* were collected in 1977, making it "The Year of *Psorophora*" in Evansville.

TABLE 4. *Larval mosquito associations found in Vanderburgh County, IN during 1974 through 1979 larval surveys.*

ASSOCIATED SPECIES	No. of associations	Ae. sollicitans	Ae. sticticus	Ae. triseriatus	Ae. vexans	An. punctipennis	An. quadrimaculatus	Cx. erraticus	Cx. pipiens complex	Cx. restuans	Cx. salinarius	Cx. tarsalis	Cx. territans	Cs. inornata	Ps. ciliata	Ps. columbiae	Ps. cyanescens	Ps. ferox	Ps. howardii	Ur. sapphirina
Aedes sollicitans	4					X	X		X	X										
sticticus	2					X	X													
triseriatus	4				X				X				X			X				
vexans	14			X		X	X		X	X	X	X	X	X	X	X	X	X	X	
Anopheles punctipennis	11	X	X		X		X		X	X	X	X	X		X	X				
quadrimaculatus	11	X	X		X	X		X	X	X	X	X	X			X				
Culex erraticus	1						X													
pipiens complex	14	X		X	X	X	X			X	X	X	X	X	X	X		X		X
restuans	10	X			X	X	X		X			X	X	X	X	X				
salinarius	4				X	X	X		X											
tarsalis	8				X	X	X		X	X			X			X	X			
territans	8			X	X	X	X		X	X		X				X				
Culiseta inornata	3				X				X	X										
Psorophora ciliata	6				X	X			X	X						X		X		
columbiae	10			X	X	X	X		X	X		X	X		X		X			
cyanescens	3				X							X				X				
ferox	3				X				X						X					
howardii	1				X															
Uranotaenia sapphirina	2								X	X										

All the 15 species reported in Siverly's 1972 work, were collected at least once during the larval survey. Several Biting Collections yielded adults of some of these species, also. (*Ae. canadensis, Ae. stimulans, Ae. trivittatus, Ae. triseriatus, Ps. ciliata, Ps. cyanescens*, and *Ps. ferox*) to name a few.

Among the 25 species collected, besides, *Culex pipiens* complex, several vectors of mosquito-borne diseases were identified. *An. quadrimaculatus*, the major vec-

tor of Malaira, and *Cx. tarsalis*, the major vector of Western Equine Encephalitis, were collected each year of the study. *Ae. aegypti*, a major vector of Yellow Fever and Dengue, was discovered in two out of the six years. *Ae. triseriatus*, the vector of the La Crosse virus of the California Encephalitis Group, was also found in the county at several sites.

In light of the presence of several vectors of mosquito-borne disease in the midst of a large human population it would be advisable for the area to have an ongoing survey of mosquitoes in the county.

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Insects and Other Arthropods of Economic Importance in Indiana During 1982

Robert W. Meyer
Department of Entomology
Purdue University, West Lafayette, Indiana 47907

Introduction

Weather conditions in 1982 were nearly ideal for most crops in Indiana. Spring moisture conditions were such that planting proceeded at a sometimes record pace, and rains thereafter were generally timely and sufficient. The fact that for the first time in recorded history June was cooler than May seems not to have adversely affected crop production - which will probably be at record or near record levels. A cool June, a cold winter (February temperatures, for instance, averaged 7° F below normal in some parts of the state) and a record-breaking cold snap in April (3° F in Kentland) may in some instances have adversely affected the insects. Whatever the cause, disruptive insect activity was at average or less than average levels.

Corn and Small Grains

A record 90% of the corn had been planted by 16 May, and a record 50% had emerged by that day. It grew at record speed, and by 11 July 20% had silked, another record. Even in the EC (East Central) and SE districts, which because of heavy soils are usually planted late, corn planting was completed almost as early as in the rest of the state. Nearly all was safe from frost by the time of the first freeze.

An estimated 40% of the corn was treated to prevent losses caused by the feeding of larvae of the western (*Diabrotica virgifera*) and northern (*D. longicornis*) corn rootworms, about as much in the southern as in the northern half of the state. At about $10/acre, that's about $25,000,000. Damage due to this insect was to some extent offset by the abundant moisture available to the plant, in untreated, infested fields. No economic silk feeding by adults was observed.

The 1st, first-instar, larva collected in Tippecanoe (WC) Co. was taken 28 May, the 1st second, by 8 June, thirds by 15 June and pupae by 29 June, in research plots regularly sampled. Adults were collected 25 June in Bartholomew (C) Co., and 30 June as far north as LaPorte (NW) Co. Adult westerns averaged 0.72/stalk statewide in an August survey, about double the 1981 population. Counties on the western side of the state averaged about 0.5/stalk, counties in the NC, C and SC districts averaged 0.65, and eastern counties 1.0, with the EC averaging 1.54, the highest in the state. Northern corn rootworm adults averaged 0.15/stalk statewide, ranged by district from 0.07 to 0.28/stalk.

A total of 17,366 adults of the two species was collected on 10 sticky traps in a Tippecanoe Co. treated cornfield between 11 June and 23 September, a figure comparable to those of 1977 and 1979, other high years. The ratio of western to northern: 79:21. Last year the ratio was 94:6.

The overwintering population (Fall, 1981) of European corn borer (*Ostrinia nubilalis*) larvae averaged 22.6/100 stalks, the lowest level in the years from 1961 to the present. By 4 May, in Daviess (SW) Co., pupation in above-ground corn stalks had begun. (In 1981 pupation in the same field was almost complete by 4 May.) The first BL (blacklight) trap catch in a Shelby (C) Co. trap of a spring

adult corn borer occurred 15 May, and catches of first generation moths peaked by 30 May at both ends of the state, when corn averaged 30 cm in height. Second generation moths peaked in numbers at the end of July in a Lawrence (SC) Co. BL trap, the first week in August in LaGrange (NE) Co., by which time about half the corn had reached the dough stage. The flight period as indicated by BL traps for second generation moths lasted through August in weather that in balance should have favored oviposition and establishment, but the insect was not able to increase its numbers through the two generations a great deal. By the time of the fall survey (September- early October) the state average for late instar larvae was only 30.5/100 stalks. (The 20-year average is 61.8.) The northern districts together with the C district had the highest averages, from 40-50 larvae/100 stalks. The highest single field average was 550 borers/100 stalks, in a Bartholomew (C) Co. field. Mechanical damage by larval feeding had resulted in only 0.2% ear loss by the time of the survey, down from 1.2% last year.

Corn leaf aphids (*Rhopalosiphum maidis*) or evidence of their once having been present were found on only 24% of the stalks in 300 fields surveyed in September-October, and 88% of those infestations were so light that they cannot have been responsible for yield losses. Infestations were most common in the northern 3 tiers of counties.

Ear tip feeding was minimal: of 7500 ears surveyed in September-October, only 0.7% had been damaged by birds, and 0.4% by fall armyworm (*Spodoptera frugiperda*) or corn earworm (*Heliothis zea*) larvae. The 14-year (1969-1982) average for birds is 4.5%, for larvae other than European corn borers is 3.9%. Only 6 larvae of each species were present at the time of the survey; the average is 65 corn earworms and 32 fall armyworms. Loss in each damaged ear was minor.

Leaf feeding and stem cutting insects included armyworms (*Pseudaletia unipuncta*), fall armyworms, black cutworms (*Agrotis ipsilon*), sod webworms (*Crambus* sp.) and common stalk borer (*Papaipema nebris*). All were minor in impact, most involved with fewer than 650 acres (of more than 6 million). Common stalk borers, normally damaging only at field edges, in one Warren (WC) Co. corn field were at economic levels through the whole wheat field that had been treated with herbicide prior to no-till corn planting. The sod webworms occurred in a corn field that had been in mint. Most armyworm problems were in no-till fields.

The oat bird-cherry aphid (*Rhopalosiphum padi*) was of little consequence in small grains this year, reaching a maximum only of 1/stem in SW district wheat, by mid-May. Early in July they were more likely to be present in whorls of corn than were corn leaf aphids. Later in the fall they were on the lower internodes, and 12.3% of the plants surveyed in September-October had colonies. Such infestations were by far more common in the northern half of the state than the southern.

A survey done cooperatively by the Agriculture Research Service of the USDA, The Indiana Crop Improvement Association and Purdue's Department of Entomology of 332 wheat fields in 52 counties noted an increase of Hessian fly (*Mayetiola destructor*) activity over that of 1981. This showed up in the mean percentage infestation rate (5.5%), the mean number of puparia/100 stems (7.4) and the percent of fields infested (63%), all higher than in 1981. These figures include all varieties of wheat. Wheats with the H_6 gene for resistance to the Hessian fly had infestation rates of less than 1.0%. The higher infestation rate was attributed to weather conditions optimal for emergence, and the presence of fly biotypes capable of attacking wheats with resistance attributable to other than H_6 genes.

The cereal leaf bettle (*Oulema melanopus*) was observed in a few wheat fields

in Harrison (SC) Co., at less than economic levels. Corn fields near the wheat received such large numbers of adults when the wheat ripened that damage to that crop was conspicuous. A field of oats in the same county was at or near treatment levels as well.

Forage Legumes and Soybeans

The following data on the alfalfa weevil (*Hypera postica*) are from SC district alfalfa fields. By 7 April, when alfalfa ranged from 7-14 cm, only trace feeding was observed. By 7 May, in alfalfa averaging 32 cm, an average of 48% of the plants had feeding. A week later buds appeared, the larvae had apparently passed their peak in numbers and alfalfa was beginning to mask the older damage. Very few fields needed treatment, although it was difficult to reach that decision at the time.

Potato leafhopper (*Empoasca fabae*) on the other hand was an economic problem on most of the alfalfa in the state and on most of the cuttings but the first and probably the last, late fall cutting.

The first adult Mexican bean beetle (*Epilachna varivestis*) of the season was collected in Jefferson (SE) Co. from alfalfa on 27 April, the normal time for its appearance. Developmental data which follow are based on research fields in Clay and Owen (WC) counties: in counties north and east development was slower, in counties to the south and southeast, slightly faster. First eggs on soybeans: 4 June. Peak first generation egg population: 17 June. Peak new adults of the first generation: 24 July. Peak new adults of the second generation: first week in September. A partial third generation may have reached third instar but made no contribution to the overwintering population.

Although there were probably fields in all bean growing counties in which at least some losses were incurred, damage was concentrated in the WC district from Clay and Owen counties in the south as far north as Montgomery County. The latter county in the west and Randolph (EC) Co. in the east mark the northern boundaries of the beetle in soybean fields with but one exception this year. There were a few treated fields in Wabash (NC) Co. this year. Such isolated infestations were more common last year, and less severe; they seldom persist for more than 1 year. Numbers entering hibernacula this year should be large.

At least 100,000 acres were treated for the control of the Mexican bean beetle in 1982, at a cost of $10/acre or more. Probably more than half of those acres needed treatment at the time the treatment was applied. There were also fields left untreated that suffered losses as a result; no estimate of the number of acres so affected is available.

The Mexican bean beetle was the only pest of consequence to soybeans in 1982.

Ornamentals, Forest, Shade and Fruit Trees

Thomas Mouzin, who operated pheromone traps in the Vincennes area (Knox—SW—Co.), provided the following fruit tree insect data. Codling moth (*Cydia pomonella*), obliquebanded leafroller (*Choristoneura rosaceana*), oriental fruit moth (*Grapholitha molesta*) and lesser peachtree borer (*Synanthedon pictipes*) catches were all at average levels. Redbanded leafroller (*Argyrotaenia velutinana*) counts were at about half the seven year average.

Beneficial Insects

Because adult alfalfa weevils were difficult to collect in numbers no estimate could be made of the rate of parasitization by *Microctonus aethiopoides*. Of the

4833 alfalfa weevil larvae collected in May and reared for parasite emergence, 51% were parasitized by either *Bathyplectes anurus* or *B. curculionis*. *B. anurus* was collected almost exclusively in the southern half of the state although it is known to exist as far north as the Michigan border. In the SC district it outnumbered *B. curculionis* by 6 to 1, in the SE by 19 to 1. On a statewide basis *anurus* accounted for 19%, *curculionis* for 32. Total percent parasitization, from west to east, in the northern districts was 62, 53 and 34; in the central: 40, 38 and 22; in the southern 64, 72 and 61.

Two attempts are made each year to estimate numbers of red coccinellids. Ten sticky traps in a treated field of continuous corn in Tippecanoe Co. collected 412 adult *Coleomegilla maculata* in the 105 days between 11 June and 23 September, the highest number in the 6 years that the traps have been used. The ratio between that species: *Hippodamia convergens*: *H. tredecimpunctata*: *Cycloneda sanguinea* was 75:10:1:14. This ratio has remained much the same except for 1977, when it was 27:65:3:5, and 1980, when it was 58:7:2:33.

A count is also made during the annual corn insect survey in Sep-Oct, of all coccinellids observed on 25 stalks in each of the 300 fields visited. In 1982 the count, in the same order as the above ratios, was 81, 11, 2, 1. The count (81) of *Coleomegilla maculata* was the lowest since 1977, has been as high as 276 (1981). This species was the most common in the NW corner of the state at the time of the survey.

A Comparison of Ant Faunae on Unreclaimed Stripmines in Indiana

JACK R. MUNSEE
Indiana State University, Terre Haute, Indiana 47809
and
JOHN R. SCHROCK
University of Kansas, Lawrence, Kansas 66045

Introduction

Previously, ants have not been considered indicator species characterizing successional stages. However, the collection of invertebrates on revegetating stripmines indicates that ants occur in abundance(3), exist in a large assortment of species, and the assortment changes between extremes, from barren ground to undisturbed forest(5).

If the ant composition of various stripmine sites are compared, differences will be readily noted (See Tables 1 and 2). Species changes as to kinds and numbers due to changes in vegetation, for example, become more apparent when the same sites are compared after a long time lapse as is done in this study.

Methods

Following cessation of mining in 1950, we examined an unreclaimed mining site fourteen and thirty-one years later. The research site consists of coal spoilbanks west of Clinton and south of Centenary, Vermillion County, Indiana. The rugged ridges were formed in 1949 and 1950 and left unreclaimed. The area was purchased by the Clinton Chapter of the Izaak Walton League, from which permission to do this study was obtained. The League has protected the area from further human activities.

In 1964, while most of the area was still in a bare ground and grass stage, nineteen sets of paired pitfall traps were established on the spoilbanks. These sampled ant populations in each of 19 stands selected on the basis of variations in slope and vegetation. Two additional pairs of traps sampled ants in an adjacent abandoned orchard and undisturbed forest. The pitfall traps were operated weekly through the summer. Accompanying weather data, soil analyses and vegetation tallies were made as described by Munsee(5).

In 1981, the same locations were sampled during the same calendar days, providing a direct measurement of changes at each site. The ants were sorted and identified for both years. The variation in ant populations among sites at varying stages of succession are given in Table 1 and 2. Prominence values, based on Beals' study of forest bird communities(1), were derived by multiplying the density of a species by the square root of its frequency. Density was determined by dividing the total number of individuals of a species collected at a site by the total number of sample periods. Frequency was determined by dividing the number of sample periods in which a species was trapped by the total number of sample periods.

The sites in 1964 were compared by calculating a similarity matrix based on the presence and absence of ant species. A similar matrix was calculated for the 1981 sites and dendrograms were drawn for each year, clustering the sites with the most similar ant samples.

Results

Prominence values of ants from the 1964 and 1981 studies are given in Tables

TABLE 1. *Average prominence values for ants in 1964.*

	W	A	B	C	D	E	F	G	H	I	J	K	L-O	M	N	P	Q	R	S	T
Aphaenogaster rudis rudis (Emery)	37.7	16.0	0.1	28.7	– –	70.7	0.3	0.3	55.2	– –	268.0	99.3	2.8	– –	0.2	– –	– –	0.1	0.1	41.7
Camponotus castaneus (Latreille)	5.7	4.4	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –
Crematogaster lineolata (Say)	2.5	57.6	27.0	33.6	8.1	125.7	39.7	28.6	172.3	3.6	186.6	99.0	16.2	2.4	28.3	11.7	4.4	0.4	23.3	15.3
Formica fusca Linnaeus	0.5	0.5	2.6	5.4	208.3	50.2	4.6	234.3	46.7	7.6	25.0	14.0	101.4	74.7	19.5	92.0	26.3	80.7	50.9	4.2
Iridomyrmex pruinosus analis (Andre)	– –	0.1	489.9	133.8	20.4	93.9	321.7	187.2	335.4	228.0	83.0	– –	42.4	257.0	323.6	352.1	330.8	132.3	622.1	101.8
Lasius neoniger Emery	3.0	4.2	11.4	64.3	0.3	– –	0.1	– –	5.3	0.4	0.1	0.1	17.5	0.7	0.5	– –	0.1	– –	– –	9.0
Leptothorax curvispinosus Mayr	4.8	1.5	– –	– –	– –	0.1	– –	– –	0.2	– –	18.1	29.0	0.3	0.1	– –	– –	– –	– –	– –	– –
Monomorium minimum (Buckley)	– –	– –	0.1	0.1	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	0.1	– –	– –
Paratrechina melanderi arenivaga (Wheeler)	0.1	40.0	0.7	– –	– –	1.8	1.2	0.1	25.9	– –	– –	– –	– –	– –	7.2	– –	– –	– –	– –	– –
Pheidole bicarinata bicarinata Mayr	– –	– –	185.6	0.8	22.7	163.1	– –	104.8	220.6	244.3	– –	– –	2.8	344.4	175.1	61.0	4.1	78.3	66.8	122.2
Ponera pennsylvanica Buckley	1.1	2.0	– –	0.5	0.1	0.5	– –	0.2	0.1	– –	11.3	18.5	0.5	0.1	0.1	– –	– –	– –	0.3	4.0
Smithistruma filitalpa Brown	– –	– –	0.1	– –	0.1	0.3	– –	1.0	– –	0.1	0.6	0.1	0.3	– –	0.7	– –	– –	0.4	– –	0.1
Solenopsis molesta (Say)	– –	50.6	5.3	227.9	0.7	18.6	515.7	29.4	18.3	1.4	130.0	102.2	502.9	38.5	82.5	14.5	36.8	25.9	12.1	22.7
Tapinoma sessile (Say)	1.1	12.6	0.7	55.5	– –	9.4	– –	0.2	– –	– –	– –	0.3	0.2	– –	– –	– –	– –	– –	– –	0.2

TABLE 2. *Average prominence values for ants in 1981.*

	W	A	B	C	D	E	F	G	H	I	J	K	L-O	M	N	P	Q	R	S	T
Aphaenogaster rudis rudis (Emery)	2.2	0.6	1.2	1.6	– –	0.5	1.8	2.2	2.4	6.3	0.7	1.1	1.7	1.2	0.8	2.1	8.1	2.7	1.6	0.3
Camponotus castaneus (Latreille)	0.4	0.5	0.1	0.1	0.1	– –	0.1	0.1	0.1	0.1	0.2	0.1	– –	– –	– –	0.1	0.1	0.1	0.1	0.1
Crematogaster lineolata (Say)	0.1	0.1	6.2	7.9	0.8	1.9	9.6	2.7	3.1	1.9	4.7	2.0	1.3	0.5	1.8	2.0	9.1	0.2	1.9	0.5
Formica fusca Linnaeus	0.2	0.1	0.4	0.1	14.1	0.5	3.3	0.5	0.4	0.5	2.2	0.1	0.1	0.3	1.3	0.1	17.0	0.4	3.1	0.3
Iridomyrmex pruinosus analis (Andre)	– –	– –	– –	– –	16.4	– –	– –	– –	– –	0.1	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –
Lasius neoniger Emery	0.1	– –	0.9	5.0	0.5	0.6	0.6	1.1	0.2	0.3	10.5	1.2	1.3	1.2	2.3	0.1	0.2	0.6	1.7	0.8
Leptothorax curvispinosus Mayr	0.1	0.1	– –	0.1	– –	0.3	0.3	0.4	0.4	0.1	0.1	0.3	0.1	0.6	0.1	0.3	0.6	0.2	0.1	0.1
Monomorium minimum Buckley	– –	– –	0.1	0.1	– –	– –	9.3	0.1	– –	– –	– –	– –	– –	– –	0.9	– –	– –	– –	– –	– –
Myrmecina americana Emery	– –	– –	0.1	1.0	– –	0.1	0.1	0.4	0.2	0.1	0.2	0.6	0.2	0.1	0.2	0.1	0.5	0.4	– –	0.1
Myrmica pinetorum Wheeler	0.1	0.2	0.2	0.2	– –	0.1	1.4	5.1	1.0	0.8	1.6	0.2	0.6	1.0	0.8	0.2	1.9	0.2	0.5	0.1
Myrmica spatulata Smith	10.2	6.1	2.3	9.9	0.4	0.5	0.1	7.7	2.3	2.9	6.6	6.7	12.4	7.5	5.9	5.3	6.3	8.0	12.0	12.5
Paratrechina melanderi arenivaga Wheeler	– –	– –	0.1	0.1	1.2	1.0	4.8	6.5	1.3	2.4	– –	– –	– –	1.3	1.4	0.8	0.1	0.1	– –	– –
Pheidole bicarinata bicarinata Mayr	– –	– –	0.1	– –	7.5	– –	– –	– –	– –	– –	0.1	– –	– –	– –	– –	– –	– –	– –	– –	– –
Ponera pennsylvanica Buckley	– –	– –	0.1	1.0	– –	0.2	0.3	– –	0.1	0.1	0.7	0.1	1.0	0.1	0.3	0.1	0.2	0.1	0.3	0.1
Smithistruma filitalpa Brown	– –	– –	– –	– –	– –	– –	– –	– –	0.1	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –	– –
Solenopsis molesta (Say)	– –	– –	1.1	– –	0.2	20.6	9.9	0.4	9.7	2.3	0.2	– –	0.1	0.6	3.6	4.8	3.5	0.2	3.1	0.2
Tapinoma sessile (Say)	– –	0.3	0.7	– –	0.1	0.9	1.8	4.0	1.2	1.9	0.3	0.5	0.3	2.8	0.4	0.8	1.3	0.1	1.8	0.2

1 and 2. Stand D remained essentially barren in both 1964 and 1981. Sites W and A are undisturbed forest and abandoned orchard, respectively. The other sites are intermediate successional stages of the stripmine spoilbanks. Some ant species were not collected on the barren sites while others were not found in the forest site. Ants such as *Crematogaster lineolata*, *Formica fusca* and *Aphaenogaster r. rudis* are fairly ubiquitous in 1964 and 1981. *Iridomyrmex pruinosus analis*, *Smithistruma filitalpa*, *Pheidole b. bicarinata* and *Solenopsis molesta* are greatly reduced or absent in 1981. New ants, ants with greatly increased numbers, and ants found in many new sites in 1981 include *Camponotus castaneus*, *Myrmica spatulata*, *Myrmica pinetorum*, *Myrmecina americana* and *Leptothorax curvispinosus*. However, many ants such as *Monomorium minimum*, *Paratrechina melanderi arenivaga*, *Ponera pennsylvanicus*, *Tapinoma sessile* and *Lasius neoniger* varied by sites in 1964 and shifted to other developing sites in 1981.

The 1964 dendrogram (Figure 1) demonstrates that, based on ant species, the stripmine sites are more similar to each other and to the barren site D, than they are to the two control sites. From Figure 2 it appears that by 1981 the

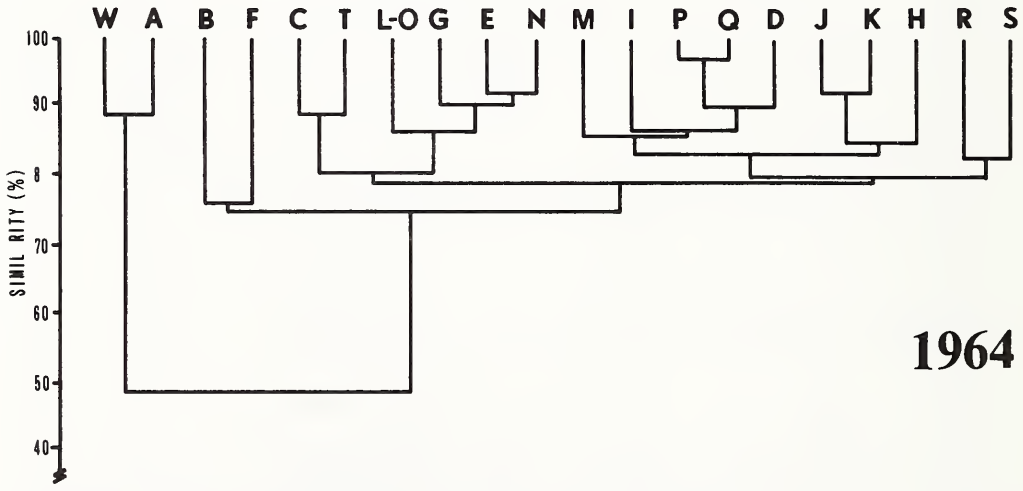

FIGURE 1. Dendrogram of study sites clustered on presence and absence of ants in 1964.

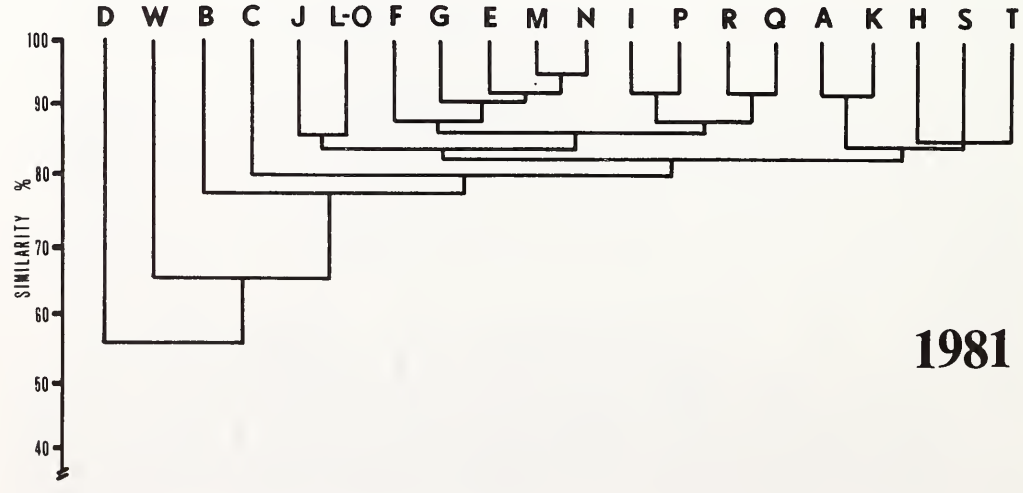

FIGURE 2. Dendrogram of study sites clustered on presence and absence of ants in 1981.

stripmined sites, except for D, are now more closely associated with the unreclaimed orchard (site A) and the forest (site W), and are not dissimilar to D.

Discussion

Although Munsee(5) found no correlation between plants and the ant fauna in 1964, a dendrogram clustering similar sites on the basis of ants alone indicated the mined sites were in an early stage of succession closer to the barren site D than to the adjacent unmined sites. Since the 1981 dendrogram based on ants shows the revegetating sites to be more similar to each other and to the unmined sites, than to the barren site D, ants alone are sufficient to indicate a general successional change.

Also, ants may not be mere passive indicators of site conditions since recent research shows several ant species are important in spreading plant seeds(2) and plant seeds buried by ants may have much better chances of survival than seeds dispersed on the surface of the stripmine spoilbanks(4). If this is the case here, some of these ant species may have a direct role in stripmine succession.

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Biogeography of Indiana Trichoptera

R. D. Waltz and W. P. McCafferty
Department of Entomology
Purdue University, West Lafayette, Indiana 47907

Introduction

The caddisflies constitute one of the largest orders of aquatic insects in North America, with approximately 1,200 species recognized North of Mexico. The first Indiana caddisfly records were reported by Banks and Ulmer independently in 1907. From 1907 to the present 89 species were reported by various workers. An additional 101 species records resulted from our study. Collections made principally over the past 11 years by McCafferty and others at Purdue University have provided the bulk of data. Specimens from non-Purdue collections were also examined, most notably from the Illinois Natural History Survey. A checklist of the 190 Indiana species has been prepared by Waltz and McCafferty (in press).

Extralimital Affinities

On a world basis the 58 known Indiana genera are more closely affiliated with the Palearctic fauna than to that of any other outside the Nearctic (Table 1). This suports data earlier presented by others, particularly Ross (7) and Chandler (2).

Table 1. *World distribution of Indiana Trichoptera Genera.*

Distribution	No. of Genera
COSMOPOLITAN	9
NEARCTIC EXCLUSIVELY	11
HOLARCTIC (PALEARCTIC & NEARCTIC) EXCLUSIVELY	14
HOLARCTIC & ORIENTAL	7
HOLARCTIC & AFROTROPICAL	1
HOLARCTIC & ORIENTAL & AUSTRALIAN	1
HOLARCTIC & ORIENTAL & AFROTROPICAL	3
HOLARCTIC & ORIENTAL & AFROTROPICAL & AUSTRALIAN	3
HOLARCTIC & ORIENTAL & AFROTROPICAL & NEOTROPICAL	2
NEARCTIC & NEOTROPICAL	5
NEARCTIC & ORIENTAL	2

Within North America, as similarly demonstrated for the Illinois Trichoptera (6), Indiana caddisflies have strong affinities with the northeastern U.S., eastern Canada, central U.S., and, to a lesser extent, the southeastern U.S. (Table 2). It

Table 2. *Distributions of Indiana Trichoptera in Other Geographic Regions. (Regions after McCafferty, 4)*

REGION	# SPECIES IN COMMON	% OF INDIANA FAUNA
Central	190/190	100.0
Eastern (Canada)	132/190	69.5
Western (Canada)	33/190	17.4
Southeastern (US)	108/190	56.8
Northeastern (US)	145/190	76.3
Northwestern (US)	46/190	24.2
Southwestern (US)	20/190	10.5

should be noted that of the species occurring in the southwestern U.S. and Indiana, ten are widespread transcontinental species (e.g., *Oecetis avara*, *O. inconspicua*). The Indiana fauna is very distinct from that of western North America. Ross (6,7,8,9,10) provided most of the present information on the origins of North American Trichoptera. He (9) concluded that major elements of Indiana and the Midwest were Tertiary.

We have found three major faunal elements of Trichoptera in Indiana. The first consists of the lotic Temperate Deciduous Forest species restricted to cool, first and second order streams. These species are most abundant in the unglaciated area, characterized as the South-Central Upland (2) from Perry, Crawford, and Harrison counties northward to an apex in Morgan County (Figure 1). This area served as a glacial refugium during the Pleistocene (2,9). Another such area exists in Parke and Montgomery counties (Turkey Run and Shades State Parks, respectively). Other widely scattered, localized areas also are characterized predominantly by this faunal element, which is represented best by the genera *Neophylax*, *Agapetus*, and *Pycnopsyche* (9). Other caddisflies such as *Potamyia flava* and the *Hydropsyche scalaris* complex share the same history of invasion but inhabit larger streams; they tend to be widely distributed and represent some of the most abundantly and commonly collected species in the state.

The second faunal element consists of widespread transcontinental species. Species such as *Oecetis inconspicua*, *O. avara*, and *Helicopsyche borealis* are reported from Mexico and Central America northward into Canada from coast to coast. Their origin is difficult to ascertain. Similar to the *H. scalaris* complex, they often occur in larger streams where they may be a dominant component of the fauna.

The third major trichopteran faunal element consists of Northern Transcontinental species that are more restricted in the state. They are most abundant in the northern third, an area with many glacial lakes and coolwater streams, and are mostly of the family Limnephilidae and Phryganeidae, although members of at least four other families are represented. Several of these species are Holarctic and of unknown origin. Some have disjunct distributions, occurring also in Perry, Harrison and Crawford counties.

Indiana species are comparable in number and composition with those reported from contiguous states (Table 3). These data indicate the presence of a rather homogeneous midwestern fauna of which Indiana is a central element.

Faunal Provinces in Indiana

Chandler (2) redefined the faunal provinces of Indiana and reviewed the earlier works on Indiana (1,3). Species found within each of these faunal provinces (Figure 1, Table 4) were compared using Sorenson's coefficient of similarity (Table 5).

TABLE 3. *Similarity of the Trichoptera Fauna of Indiana and Adjoining States.*

	#SPECIES	#GENERA	SPECIES SIMILARITY[1]		GENERIC SIMILARITY[1]	
TEST			Jaccard	Sorenson	Jaccard	Sorenson
IN	190	58	1.0	1.0	1.0	1.0
xMI	181	61	.4220	.5935	.6667	.8000
xKY	175	54	.4622	.6322	.7813	.8772
xIL	184	58	.6860	.8138	.8281	.9059
xOH	192	56	.4920	.6595	.7692	.8695

[1]Jaccard's Coefficient: a/(a + b + c); Sorenson's Coefficient: 2a/(2a + b + c); Where a = # in common (both states), b = # reported in Indiana only, c = # reported in alternate state only.

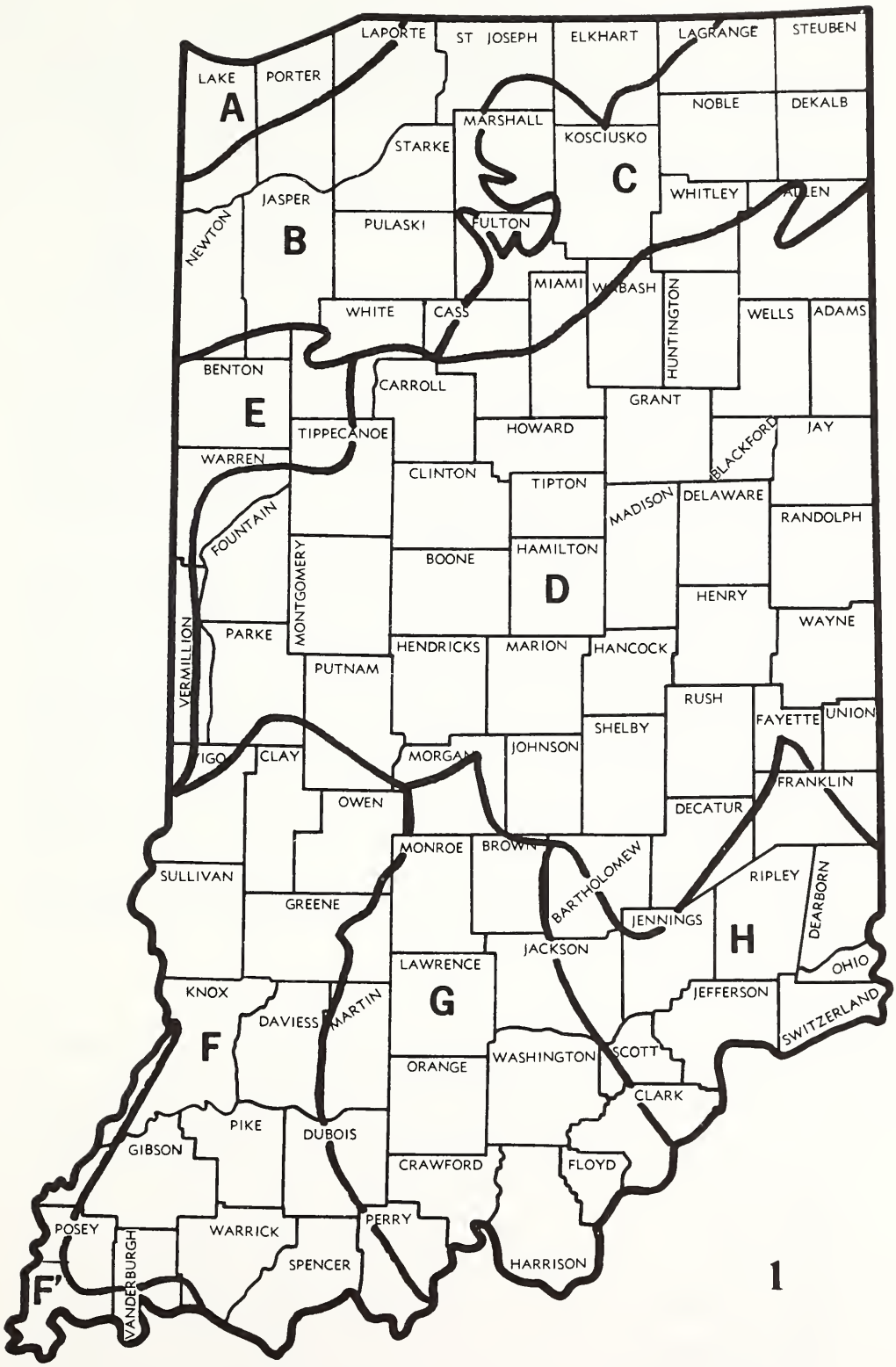

FIGURE 1. *Indiana counties and faunal provinces (after Chandler, 2). A. Dunes. B. Kankakee Sand Ridge. C. Northern Lake. D. Tipton Till Plain. E. Prairie. F. Southwestern Plains and Upland. G. South-Cental Upland. H. Southeastern Till Plain. F'. Lower Wabash Valley.*

TABLE 4. *Trichoptera of Indiana Faunal Provinces (after Chandler, 2).*

PROVINCE	# Species	# Unique
Tipton Till Plain	73	17
Southeastern Till Plain	15	2
Southwestern Plains and Upland (Inclusive of Lower Wabash Valley)	21	2
Prairie	4	3
Kankakee Sand Ridge	51	5
Northern Lake	62	13
South Central Upland	76	23
Dunes	8	2

This analysis indicates a considerable obscuring of some of the provinces that may in part be attributable to the dispersal ability of caddisflies in addition to caddisflies' dependence on aquatic rather than terrestrial habitats, which formed the basis of Chandler's regions. Our distributional data share some similarity with that presented for the Indiana Odonata (5).

The northern third of Indiana, the most heavily glaciated region, is rich in lake and marsh habitats. It isn't surprising that the two faunal regions recognized in this area (based on aquatic characters) are very similar in composition (Tables 4 and 5) as was demonstrated by Montgomery (5).

One of the most interesting areas is the South-Central Upland, which is best characterized by the presence of a distinct but diluted Appalachian fauna. We found 23 species in this area but nowhere else; this is the largest number of "area endemic" caddisflies in Indiana (Table 4). This province supports some species (e.g., *Hydatophylax argus*) that typically occur only in the Northern Lake area and Kankakee Sand Ridge in unimpacted streams (e.g., Pigeon River, LaGrange County).

Little can be said of the remaining faunal areas. The Prairie province of Indiana, although small, may prove to be faunistically unique and a source of several more records.

TABLE 5. *Intrastate Faunal Province Affinities Based on Trichoptera Composition. (Based on Sorenson's coefficient of similarity, SCS = 2a/(2a + b + c).*

	SCU	TTP	NL	KSR	SWPU	SETP	DUNES	PRAIRIE
South Central Upland (SCU)	1.0	.429	.348	.315	.247	.154	.071	.025
Tipton Till Plain (TTP)		1.0	.444	.468	.234	.250	.074	.026
Northern Lake (NL)			1.0	.584	.193	.104	.086	.030
Kankakee Sand Ridge (KSR)				1.0	.277	.090	.102	.036
Southwestern Plains & Uplands (SWPU) (Inclusive of Lower Wabash Valley)					1.0	.056	.276	.080
Southeastern Till Plain (SETP)						1.0	.087	—
Dunes							1.0	—
Prairie								1.0

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Ball State University, Munice, Indiana 47306

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Lafayette, Indiana, 47902

ABSTRACTS

Ambient Air Ozone Concentrations in Central Indiana. WILLIAM BERANEK, JR., Environmental Research Service, Indianapolis Center for Advanced Research, Indianapolis, Indiana 46202.——Marion County has been declared a nonattainment area for ozone air concentrations by the United States Environmental Protection Agency. Control measures have been instituted to reduce hydrocarbon emissions in Marion County in order to reduce the number of ozone excursions. Since ozone is created by sunlight from chemical precursors, it is important that the relative contribution of sources of all precursors be understood in order to prepare an effective control policy. Data on time, location and intensity of ozone air concentrations in central Indiana indicates that a major contribution to ozone concentrations on some critical days occurs from precursors external to the area. This evidence is presented and discussed in the context of pollution control policy.

Potential Effects of Acid Rain on the Mortality and Development of *Aedes triseriatus* **Larvae and Pupae.** KEITH R. FADELY and HORST F. SIEWART, Department of Natural Resources, Ball State University, Muncie, Indiana, 47306.——Larvae of *Ae. triseriatus* mosquitoes were collected from treeholes on the Ball State University campus. These insects were raised to adulthood under laboratory conditions. Newly hatched larvae were raised in deionized water. One hundred twenty-five instar 1 mosquitoes (group A) were placed into separate baby food jars containing deionized water. These jars were separated into five groups. The pH of the water in each group was 3, 4, 5, 6 and 7. Subsequent groups composed of 125 instar 2, 3 and 4 larvae and pupae (groups B, C, D and E respectively) were exposed to the same experimental treatment as that of group A. The effects of each of these pH levels on mortality and development was observed until the insects reached maturity or died.

Larvae in water of pH 3 experienced higher percentage of mortality than mosquitoes exposed to more basic solutions. Larvae experienced a decrease in mortality with age. Instar 4 experienced the lowest mortality. Mortality of immature *Ae. triseriatus* in groups A and B was highest during their development as instar 2 larvae. The lowest mortality occured at instar 4 among groups A, B, C and D. The duration of developmental time among larvae in pH 3, 4, 5, 6 and 7 decreased as the hydrogen ion concentration decreased. Mosquitoes in solutions of pH 3 required the greatest amount of developmental time. At pH 7, the shortest time was required. Developmental time increased with larval age as acidity increased. Instar 1 larvae required less time than any other developmental stage. The longest amount of time was required by instar 4.

A Revision of the Traditional K_2—Temperation Relationship of Stream Reaeration. ROBERT H. L. HOWE and JOHN M. HAWKINS, Institute of Advanced Environmen-

tal Health Science Research, Joint Project, West Lafayette, Indiana 47906.——The oxygenation coefficient of streams (K_2) is temperature dependent and the following equation has been used for decades to calculate the K_2 value at higher temperature levels by its known value at 20°C:

$$K_{2(T)} = K_{2(20)} \times \theta^{T-20} \text{ - - - - - Eq.(1)}$$

where: $K_{2(T)}$ = oxygenation coefficient at T°C.

$K_{2(20)}$ = oxygenation coefficient at 20°C.

θ = temperature coefficient

The value of θ is greater than 1.0 (1.01 - 1.05) thus equation (1) delineates an increasing K_2 value at higher temperature.

Recently, Howe (1977) proposed that the K_2 value decreases as the liquid temperature increases. Based on his laboratory data, the following modification of the K_2—temperature relationship was proposed:

$$K_{2(T)} = K_{2(20)}/\theta^{T-20} \text{ - - - - - Eq.(2)}$$

and $\theta = 1.125$

our recent results have shown that the K_2—temperature relationship is dependent on the magnitude of K_2. When the K_2 value is below 40 day^{-1}, it decreases at increasing liquid temperatures while other factors remain constant. Howe's modified equation seems to be valid for oxygenation conditions of relatively low air flow rates or power input for aeration.

The Air Discharge per Manufacturing Employee in Indiana. STUART L. IRWIN AND WILLIAM BERANEK, JR., Environmental Research Service, Indianapolis Center for Advanced Research, Indianapolis, Indiana 46202.——Several locations in Indiana have ambient air concentrations of sulfur dioxide and particulate which exceed the primary standard established by the United States Environmental Protection Agency. Combustion of coal and oil by manufacturers in Indiana is responsible for most of the point source discharge of sulfur dioxide and particulates. Since the U.S. EPA requires economic development to be accomplished without deterioration of air quality, Indiana policymakers must be able to predict the impact of air pollutant discharges from the anticipated increase in manufacturing activity.

To predict future air pollution in communities in Indiana, it is necessary to estimate the growth in the manufacturing sector by industry type and by facility size. Different industry types have different discharge characteristics. Different facility sizes have different control requirements and different impacts on air quality. We have calculated a general ratio of pollution per employee for Indiana for the industry-types projected to expand in Indiana. We have compared the calculated emission increase with actual emissions by industry in Indianapolis. This ratio can be used by air quality planners along with economic projections to estimate changes in overall air quality.

Simulated Chemical Weathering of Coal and Overburden Materials. PATRICK J. SULLIVAN, Department of Natural Resources, Ball State University, Muncie, Indiana 47306.——One Pennsylvanian coal, one Pennsylvanian gray shale and a Cretaceous red sand from the Kellerman mine in Tuscaloosa, Alabama were leached in soxhlet extractors at 25°C for 144 hours. Identical samples were leached once a week for 11 weeks using a humidity cell at approximately 25°C. Leachate collected

from the soxhlet and humidity cell apparatus was analyzed for pH, EC, SO_4, Fe, Mn and total acidity.

The data indicate that concentrations of SO_4, Fe, and Mn in the leachate (both the soxhlet and humidity cell) are greater from the coal and the shale greater than the red sand. This is primarily the result of pyrite and soluble phases in the coal. A comparison of the leachate characteristics of the soxhlet and humidity cell showed that the soxhlet removed 50% more total Fe, Mn, and SO_4 for the complete leaching cycle. Thus, the origin of leachate acidity and weathering products in the humidity cell could not be distinguished between the soluble mineral phases and weathering reactions.

The results of this experiment suggest that the use of the soxhlet extractor for leaching coal materials may produce a faster and more accurate representation of the order of chemical weathering than humidity cell.

Analysis of Organic Compounds in Water Samples by Gas Chromatography—Mass Spectrometry

Joseph R. Siefker and Nor'ini B. Sapuan
Department of Chemistry
Indiana State Univerity
Terre Haute, Indiana 47809

Introduction

Steadily rising industrial output and rapid population growth have deteriorated the quality of the water environment in many of the nation's lakes, rivers and ground water. In recent years widespread interest has been focused on the determination of trace organic compounds in water. This is primarily due to the evidence that some of the organic compounds that are continuously being discharged into the nation's streams and rivers are harmful.

The purpose of this study was to determine the presence of volatile organic compounds in selected water samples. Samples were collected from places where a certain degree of water pollution was suspected. These samples included well water from several places in Indiana; Wabash River water; Deming, Dobbs, and Fowler Park lake water; Sewage Treatment Plant water; and tap water.

Experimental

Water samples were analyzed for volatile organic compounds by a purge and trap method. This method utilizes gas bubbles to transfer organic compounds from the aqueous phase to gaseous phase by bubbling helium gas through the water samples. These organic compounds are then transported by the helium gas into a porous polymeric trap where the trap adsorbent retards and concentrates the purged compounds while the helium gas is vented. The compounds are then transferred into the gas chromatograph for analysis by backflushing the trap at elevated temperature (180°C).

The purge and trap equipment consisted of a purging device, trap column, and desorber. The purging device was a 500 ml gas-washing bottle equipped with a ground glass joint and a 20 mm medium fritted disc at the end of the glass tube to disperse the purging gas. The inlet and exit ports were made of a ¼" O.D. glass tubing which permitted leak-free removable connection with tygon tubing. At the inlet end, the tygon tubing was connected to the helium gas line by a ¼" O.D. copper tubing. At the exit end, the tubing was attached to a trap column.

The trap columns were made of ¼" O.D. stainless steel tubing of about 140 mm length. These columns were also provided with exchangeable Swagelok fittings, enabling the columns to be connected to gas line copper tubing during thermal desorption and conditioning of the traps. These trap columns were packed with glass wool at both ends. The central portion contained Tenax-GC (2,6-diphenylene oxide) and silica gel packings.

The desorber consisted of a heating mantel which was regulated by a temperature controller unit. Optimum heating setting of this controller was found by experimentation. For quantitative desorption it was necessary to heat the trap column as quickly as possible to 180°C without exceeding the thermal stability limit of the column packing.

The GC-MS system consisted of a Hewlett-Packard model 5712A gas liquid

chromatograph and HP 5930A quadrupole mass spectrometer. The gas chromatograph was equipped with a model 5705A thermal conductivity detector and a 5702A model temperature controller. The columns used were matched 6 ft x 1/8 in. O.D. stainless steel, packed with methyl vinyl siloxane, UCW-98, on 80-100 mesh Chromosorb G acid washed with dimethyl dichlorosilane. The recorder used was a Hewlett-Packard 7127A recorder set at 1 inch/min.

The gas chromatograph operating conditions were as follows:

Helium --- 40 psi	Column Oven Temperature:
Nitrogen --- 50 psi	Initial --- 60°C for 2 min
Injection Port Temperature---250°C	Rate --- 8°C/min
Detector Temperature ---250°C	Final ---200°C for 16 min
Sensitivity --- 5	
Attenuation --- 2	

The molecular separator used in this system between the gas chromatograph and mass spectrometer was a silicone semi-permeable membrane.

The mass spectrometer operating parameters were as follows:

Electron Energy	--- 70V	Gain	--- 3
Emission Current	--- .3 mA	Mass Filter Temperature	--- 100°C
Ion Focus	--- 40 V	Ion Source Temperature	--- 250°C
Repeller Potential	--- 28 V	Scan Speed	--- 20
Analyzer Pressure	--- 1.2×10^{-6}	Bandwidth	--- 73
Mass Range Setting	--- 30-250	Volt/Div	--- 0.2

Prior to daily use, the Tenax traps were conditioned by placing the trap inside a heating mantle that was regulated to heat constantly at 180°C. The exit end of the trap was connected to the helium gas line by a Swagelok teflon fitting and a ¼" O.D. copper tubing. The traps were conditioned for 30 minutes with 10 psi helium passing through. The traps were cooled and were ready for purging and trapping. A 300 ml sample was poured into the purging device. The Tenax trap inlet was attached directly to the purging device exit using tygon tubing. The helium gas line was connected to the device inlet using Swagelok nylon fittings, ¼" copper tubing, and tygon tubing. 20 psi helium gas was bubbled through the sample for 15 minutes to transfer the volatile organics from the sample to the trap, where the organics were adsorbed by the trap packing. The trap was then removed from the device, sealed with caps, and placed in a 4°C cooler until it was used.

The next step was desorption and analysis. The gas chromatograph was cooled to 30°C by leaving the oven door open. The caps were removed from the Tenax trap containing the sample, and a 22 gauge needle was attached to the end of the trap. The trap was then inserted into a heating mantle that had been preheated to 180°C and regulated to stay at that temperature. The other end of the trap was connected to the helium gas line by a nylon Swagelok fitting. The trap needle was immediately inserted into the injection port septum of the gas chromatograph and the hot trap was backflushed with 40 psi helium for 3 minutes.

After desorption and sample tansfer was complete, the needle was removed from the injection port, the oven door was closed, and the gas chromatograph column oven was rapidly heated to the initial operating temperature of 60°C. This temperature was maintained for 2 minutes; then the oven was heated at a rate of 8°C per minute to a final temperature of 200°C. The oven was kept isothermally at 200°C for 16 minutes. As each organic compound was eluted from the column, a mass spectrum was taken.

Results and Discussion

Some well water samples were taken from Seymour, Indiana. For a period of twelve years, more than 60,000 drums of chemical wastes were stored in Seymour's Freeman Field Industrial Park by the Seymour Recycling Corporation. Most of these drums were rusty, and chemical wastes leaked out of the corroded drums. Some residents feared that chemicals might have seeped through the ground to pollute the ground water beneath the park and to contaminate nearby private wells as well (2). Removal of the drums was proceeding during 1982(1).

During 1980 and 1981, samples were collected from wells within one to two miles of the dump site. A definitive spectrum for toluene was obtained from some of the samples. Other spectra gave no definite results although 1-methyl naphthalene, 1,2-dimethyl naphthalene, and 1-1-biphenyl were possibly indicated. Other spectra obtained from the samples could not be matched with reference spectra.

During 1981, Deming Park Lake, Dobbs Park Lake, and Fowler Park Lake—all in the Terre Haute area—were sampled. Toluene and 1,2-dichloropropene were found to be present in the Deming Park Lake sample. Benzeneacetonitrile methylene was indicated in a Dobbs Park Lake sample. Other spectra could not be matched with reference spectra.

The mass spectra from some samples of Wabash River water showed the presence of 1-ethyl-3-methyl-benzene. Other spectra obtained from Wabash River water could not be matched with reference spectra.

Samples from the Terre Haute Wastewater Plant gave spectra indicative of 3,3-dichloropropene, 1,2-dichlorobenzene, toluene, 1,4-dimethylbenzene, tetrachloroethane, 1-(1-propynyl)-cyclohexane, and other compounds that could not be identified.

Terre Haute tap water was also sampled. Most of the samples gave a few spectra with small peaks that could not be identified. However, one sample gave spectra quite indicative of dibromomethane and dibromochloromethane.

The conclusion was reached that this method allows detection of volatile organic compounds in water samples at the parts-per-billion level, but that matching of spectra with reference spectra is difficult and may sometimes be complicated because two or more compounds may elute from the gas chromatograph at nearly the same time. Nevertheless, the method is very sensitive and some spectra are quite definitive.

Acknowledgment

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The Influence of a Combined Sewage Outfall on the Chemical and Biological Quality of Salt Creek, Porter County, Indiana

RICHARD L. WHITMAN
Department of Biology, Indiana University-Northwest
Gary, Indiana 46408

Introduction

The Salt Creek catchment represents an important NW Indiana watershed strongly influencing the socio-economic and recreational resources of Porter County. Intensive land use characterizes the areas adjacent to Salt Creek and today only about 11% of its watershed remains undisturbed (2). Although riparian lands have traditionally been dedicated to agriculture (54% in 1970), croplands are rapidly being replaced by urban and suburban expansion. Zoning officials have now reserved about 65% of the watershed for residential development, 20% to commercial ventures and only 18% to agricultural land use. There are presently 11 officially recognized domestic sewage sources discharging into Salt Creek or its tributaries. The potential impacts of sewage effluent on water and biological quality may be substantial, especially in the stream's upper reaches where urban development is concentrated and stream discharge is characteristically low.

The Indiana Dept. of Natural Resources (2) has estimated that Salt Creek represents 33% of the available salmon spawning tributaries in Indiana; 75% when the receiving waters of the East Little Calumet are included. In spite of this apparent fishery potential, efforts to sustain a viable population of Coho salmon in the stream, either as a "put and take" or as a naturally reproducing fishery, have been disappointing. Locally, well publicized fish kills in 1972, 1973, and 1979 served to highlight suspected water quality problems of Salt Creek. Studies by Cole and Assoc. (1), Ledet (5) and TenEch (8) inspected the water quality of Salt Creek, but were unable to agree on either the major sources of pollution, or its impact on recreational fishing. These studies, nevertheless, did agree that further investigation was needed before the environmental status of Salt Creek and the impact of combined sewage outfalls (CSO) could be fully assessed.

This paper described some of the limnological characteristics of Salt Creek and examines the impact of major sewage outfalls on stream sediment, water and biological quality.

Description of Study Area

Salt Creek originates as cool springs approximately 2.5 km due east of State Route 49 and about 3.8 km south of Valparaiso in Porter Co., Ind. (Figure 1). The stream meanders for about 39 km in a generally northward direction to its confluence with the east branch of the Little Calumet River, north of Portage, Ind. or about 244 m upstream of the Samuelson Road crossing. The Salt Creek watershed drains a 197.4 km^2 area and has 86.6 km of channel. Major tributaries which empty into Salt Creek include: Dammon Run, Swanson, Lamporte and Clark's Ditch; Squirrel, Pepper and Sager Creeks. Ledet (5) reported a mean summer discharge of 1.17 cu. m/sec for Salt Creek at its confluence with the East Little Calumet River, although Whitman (13) reported mean summer discharges between 1.2 and 4.69 cu. m/sec at a station which was 1.5 km upstream of the latter location. According to the DNR (2), organic loading of Salt Creek originates from 4 major sources: Valparaiso, S. Haven, Robinwood Subdivision and Porter County

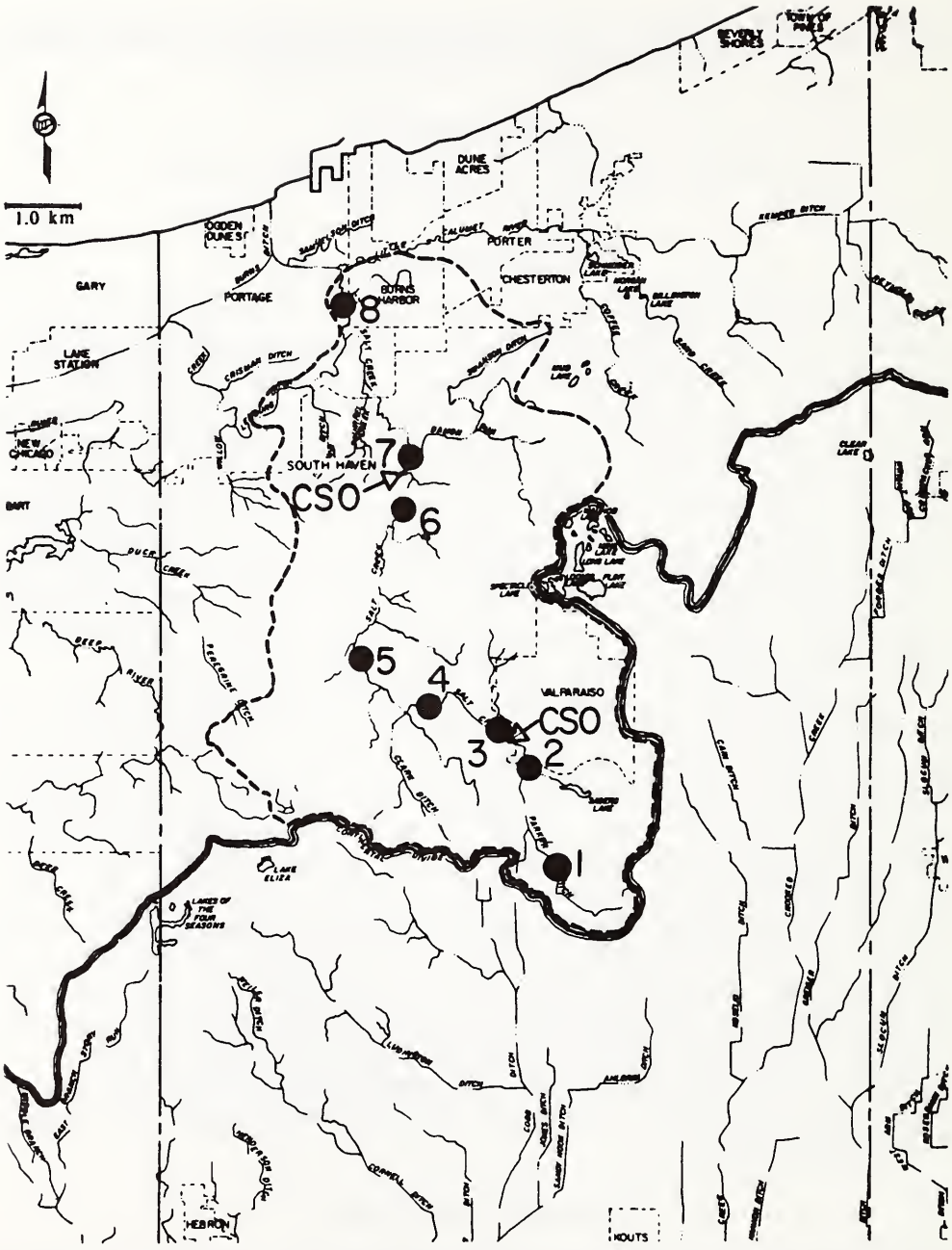

FIGURE 1. *Map of the Salt Creek watershed showing major combined sewage outfalls and sampling stations.*

Home. Other sewage treatment outfalls operate at subcapacity levels and presumably are more minor in impact. An area map showing sampling station locations is given in Figure 1.

Materials and Methods

Water, sediment and benthos samples were obtained along a three point transect representing lateral and center flow regimes. Eight stations were selected and sampled during July, Aug. and Sept. 1980 and Apr. 1981. Samples were collected, preserved and held according to conditions outlined by the U.S. Environmental Protection Agency (9, 11).

Dissolved oxygen, specific conductance, pH, temperature, and discharge were determined at the sampling sites. Dissolved oxygen was analyzed using the modified azide Winkler technique, specific conductance by a Lectro MHO meter, pH and temperature by a portable digital pH meter, and stream velocity by a General Oceanic Digital flow meter. Both velocity measurements and water quality samplings were performed at 60% of the sampling depth. Stream cross sectional area was determined at 1 m intervals across the stream transect at the lower 7 stations and every 0.5 m intervals at the uppermost station.

Laboratory water quality analyses included total ammonia nitrogen, total phosphates, and total suspended solids (non-filtratable residues at 105°C). Ammonia nitrogen was determined by subjecting 300 ml of stream water to Keldahl distillation and spectrophotometric analysis of the Nesslerized distillate. Nitrate nitrogen was determined using a Corning ion specific electrode. Total phosphate was determined by persulfate digestion and stannous chloride analysis. Total suspended solids (TSS) was assessed by filtering 200 ml of sample through a Whatman GF-1 glass fiber filter and then weighing the dried filter.

Sediment samples were obtained by inserting a 5 cm (I.D.) acrylic coring tube into the substrate to a 30 cm sediment depth. The tube's exposed end was then plugged, and the contents retrieved, measured and visually described. Cores were then transferred to Whirlpak bags, chilled and transported to the laboratory for immediate analysis. Samples were carefully mixed with a glass rod and screened through a #30 U.S. standard sieve before subsampling and analysis. These samples were then analyzed for ash-free dry weight and sediment oxygen demand. Sediment ash-free dry weight was obtained from a 10 gm subsamble and ignited in a muffler furnace at 550°C for 2 hrs. The resultant weight loss was taken as an estimate of organic content. Sediment oxygen demand was determined on 3 gms. of sediment using Gilson Respirometry. Reaction flasks were placed in water adjusted to 20°C and allowed to incubate for at least 4 hrs. at an oscillation rate of 30 cm/sec. Flasks were wrapped in aluminum foil and kept in the dark during the experiment in order to suppress autotrophic oxygen production. Differential gas readings were taken every 20 min. and later adjusted to standard temperature and pressure. The resultant slopes of oxygen uptake over time (as determined by regression analysis) were used to determine oxygen uptake rates. Except for the above modification, oxygen uptake analysis followed Umbreit et al. (12).

Triplicate benthos samples were taken with a standard Ekman grab sampler just upstream of the water and sediment quality transects. Samples were washed in a U.S. standard #30 mesh sieve and the contents transfered to quart jars and preserved in 10% formalin. Five percent rose bengal stain was added to facilitate the separation of the benthos from the debris. When practical, organisms were identified to genus.

Further details on methodology, materials, sampling techniques and station description may be found in Whitman (13).

Results and Discussion

Mean water quality trends are displayed in Figures 2a-f. Dissolved oxygen (DO), specific conductance, pH, total ammonia nitrogen, nitrate nitrogen, and total phosphate indicate that the Valparaiso CSO had a major impact on the ecology of Salt Creek. This phenomenon is especially evident during low flow conditions. Although DO during Aug. and Sept. was generally high just below the Valparaiso CSO (Station 3), DO decreased rapidly downstream. Streter and Phelps' calculations, after Linsley and Franzini (6), and empirical observations indicate that the

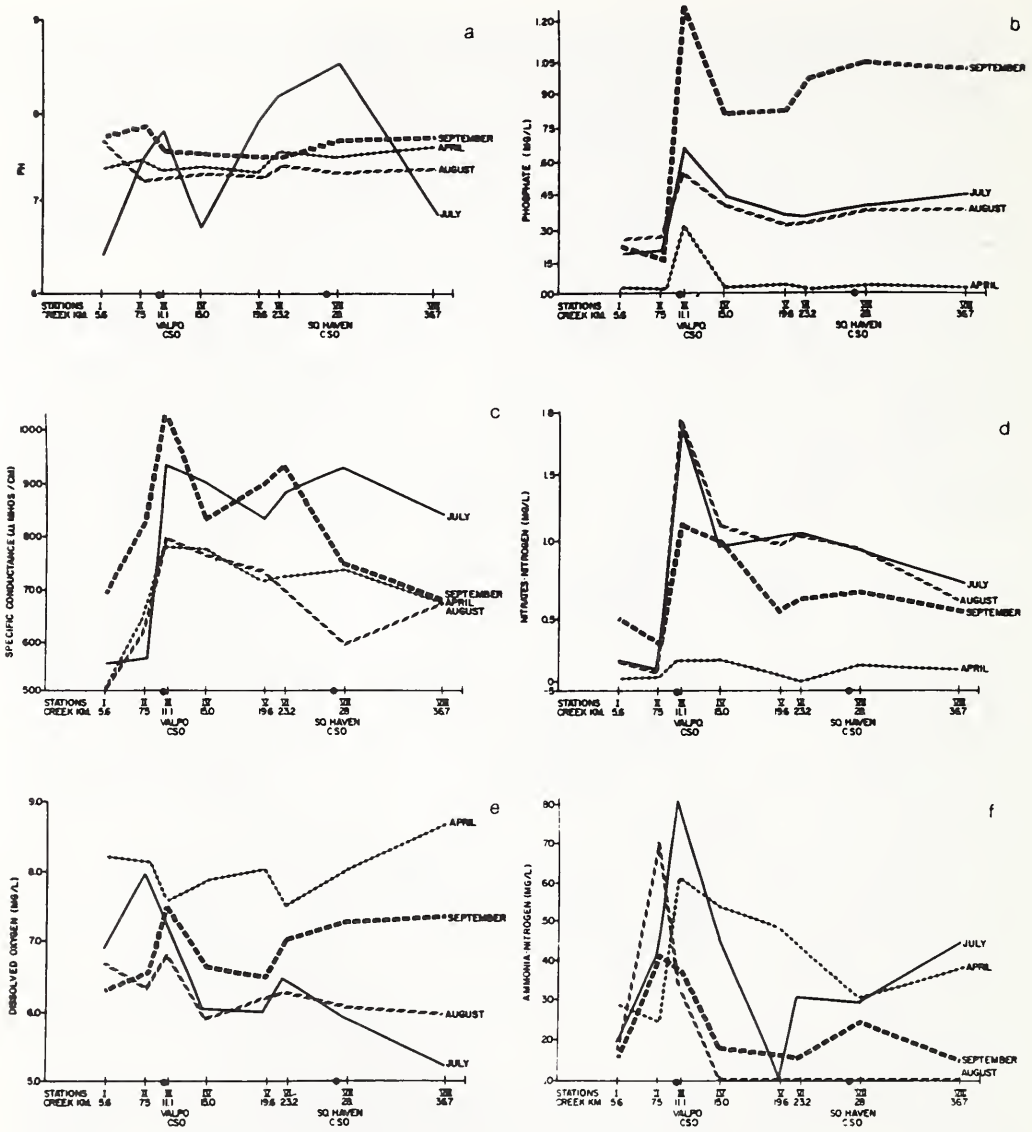

FIGURE 2a-f. *Mean pH, a; total phosphates, b; specific conductance, c; nitrates nitrogen, d; dissolved oxygen, e; and ammonia nitrogen, f; by stream kilometer for July, Aug. and Sept. 1980 and April 1981.*

oxygen sag point occurred between Stations 4 and 5. DO observations made in April suggest an oxygen balance quite unlike those seen during the warmer months. During the April outing, oxygen actually increased at Stations 4 and 5, while slightly decreasing at Stations 3 and 7. It is conceivable that under increased spring discharge the oxygen content of the stream is effectively lowered by the effluents from the Valparaiso and S. Haven combined sewage outfalls even after aeration. Cooler water, decreased biological activity, increased stream turbulence and discharge may have accounted for the rapid reoxygenation of water below Stations 3 and 6.

Figure 2c clearly shows a sudden rise in specific conductance at Station 3 followed by partial recovery to near pre-impacted levels some 37 km downstream during the Sept., Apr. and Aug. observations. July observations, in contrast, indicate little recovery of the stream from ionic loading. Low flows in July may

have helped maintain point source effects. Similar relationships apparently exist for stream pH during July. During this month, pH dropped dramatically below the Valparaiso and S. Haven outfall. Other sampling periods show no significant decrease in pH along the stream ($p = 0.05$). Decreases in pH are likely due to carbon dioxide buildup from bacterial respiration of CSO derived organics. Pearson Correlation Coefficients indicate that pH is inversely correlated with sediment organics ($p = 0.008$), sediment oxygen demand ($p = 0.017$), and positively correlated with nitrate ($p = 0.038$), chloride ($p = 0.017$), and sulfate ($p = 0.018$).

Increased in ammonia nitrogen were observed at Station 2 in Aug., Sept. and Apr. and at station 3 in July and Apr. Ammonia decreased rapidly below the Valparaiso point source. A strong smell of sewage was noted during several of the preliminary visits at Station 2. The source of the odor has not been concretely identified and other water quality parameters have not generally been supportive of sewage releases at or above this station. Conversely, nitrate increased some 5 fold between Stations 2 and 3 during July and Aug. Both ammonia and nitrate increased only slightly (if at all) at the S. Haven CSO. Figure 2b shows that total phosphate greatly increased at Station 3 followed by some recovery at downstream stations. Unfortunately pre-impacted levels were not regained during decreased flows conditions of July, Aug. and Sept. The increased flow during April presumably aided the stream's recovery from phosphorus loading. Little, if any, increase in total phosphate was indicated for stations below the S. Haven CSO.

Figures 3a-d present mean values for sediment ash-free dry weight, clorophyll a extracted from periphyton growing on glass slides as described by Patricks et al. (7), sediment oxygen demand, and benthic species diversity.

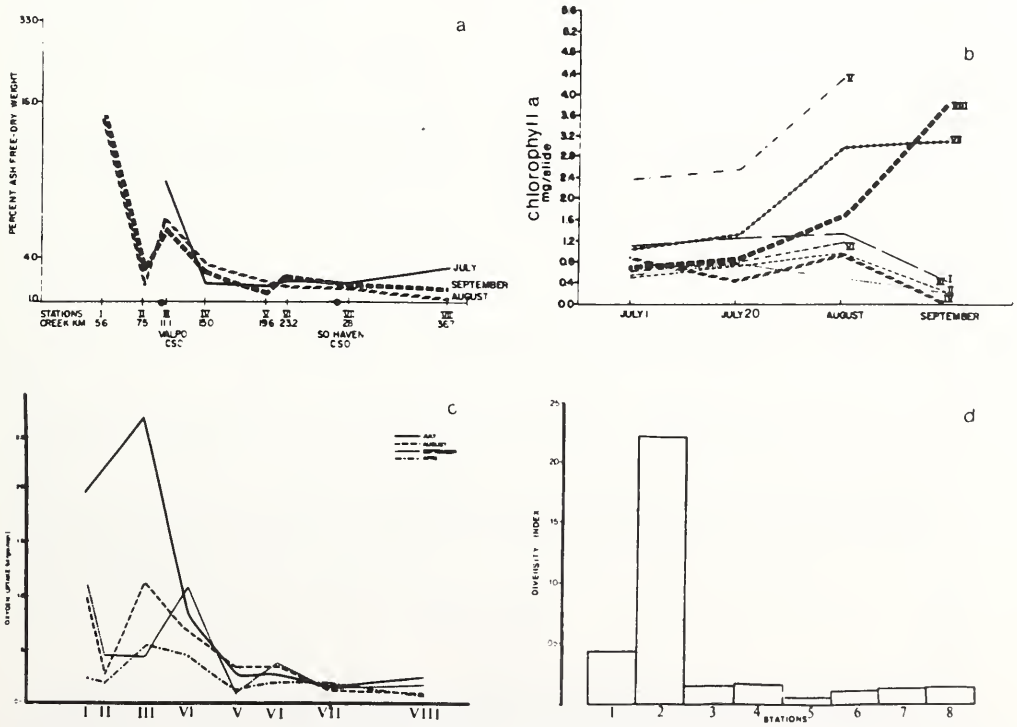

FIGURE 3a-d. *3a. Mean percent ash-free dry weight by stream kilometer for July, Aug. and Sept. 1980, and April 1981. 3b. Clorophyll a content per individual slide removed at intervals between July 1 and Sept. 30. 3c. Mean sediment oxygen demand. 3d. Overall mean species diversity of benthos taken during the study period.*

Sediments collected from Station 1 ranked highest in ash-free dry weight and were significantly higher than all, but Station 3 ($p = 0.05$). Sediment organics from Station 1 largely consisted of detritus-laden silts which presumably originated from a combination of: 1) detrital input from marshes that surround the creek's upper reaches, 2) leaf litter from the heavy canopy of vegetation along this stream stretch, and 3) possible organic particulate containing runoff from rural land use. Nutrient levels as well as periphyton productivity were also limited at the uppermost station. In contrast, organics taken from Station 3 consisted mostly of sludge deposited along the margins and banks of the stream. Periods of moderate to fast flows tended to expose the natural glacial till in the creek's center.

Results from oxygen uptake experiments (Figure 3c) support the conclusion that the Valparaiso CSO adversely impacts Salt Creek. In 3 of the 4 outings, sediments from Station 3 had the highest oxygen demand. Oxygen demand at this station was quite variable since samples from the creek's center were often composed of gravel and had a typically low organic content, unlike the sludge laden lateral portions. Earlier rains during Sept. exported sludge from Station 3 to Station 4 where it became trapped between dense stands of *Elodea*. Sludge resuspension exposed the underlying gravel and consequently lowered the substrate's oxygen demand.

Diurnal observations taken from pre- to post- rain storm events indicate that overloading of the Valparaiso CSO occurred even with moderate rainfall and resulted in anaerobic or near anaerobic conditions downstream (Figures 4). This phenomenon was observed in both the July and August diurnals. Given sufficient seasonal flow, some recovery is evident at some point downstream from the outfall. A case in point are the diurnals taken between 1850 hrs of July 25

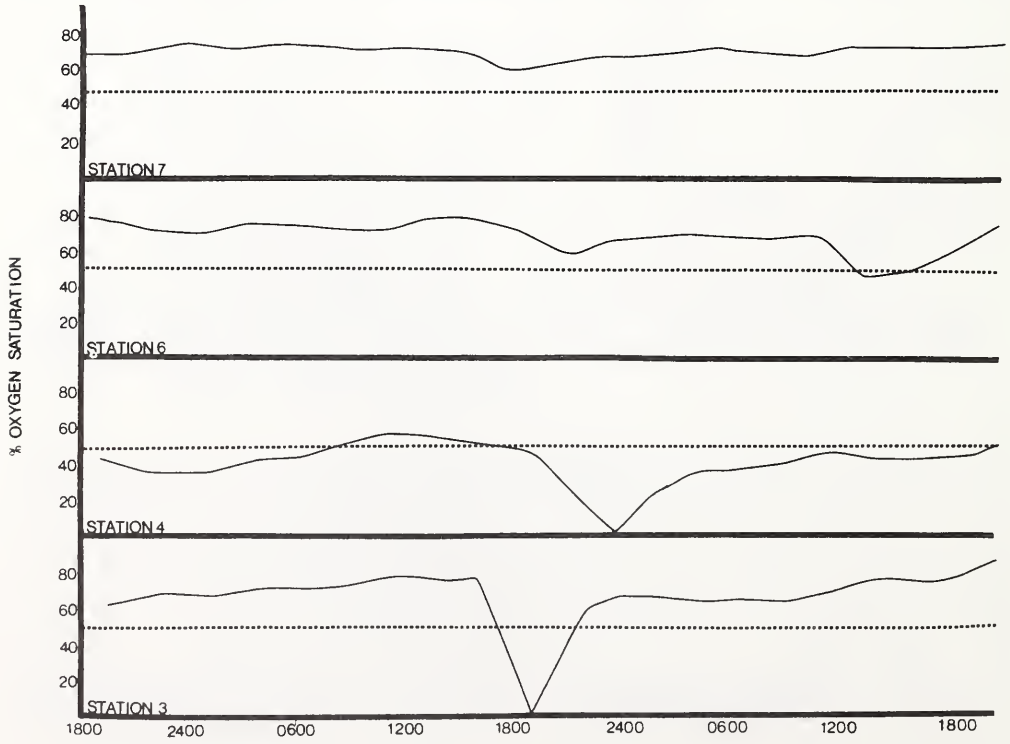

FIGURE 4. *Diurnal dissolved oxygen between 1815 hrs, July 25 to 1805 hrs July 26, 1980 at Salt Creek Stations 3, 4, 6, and 7. Station 2 was also monitored during Aug., Sept. and April diurnals.*

and 1805 hrs of July 26. During this diurnal, 0.51 cm of rain fell between 1500 and 1640 hrs of July 26 (maximum intensity occurred at 1610 hrs). DO readings at Station 3 fell from 7.2 ppm at 1605 to 0.25 ppm at 1903. Other stations sampled during this period remained between 4.7 and 6.6 ppm, reflecting more moderate decreases in DO over the proceeding observations. These latter depressions in DO are likely best explained by non-point source effects such as marsh or ground water displacement or surface runoff with appreciable immediate oxygen demands. DO at Station 3 quickly returned to near preimpacted levels at 2150 hrs or about 2.75 hrs after deoxygenation. The plume of poorly oxygenated water was detected as it passed Station 4 (4.3 km downstream of the Valparaiso CSO) at 2135 hrs with oxygen dropping from 4.7 to 1.9 ppm. Complete anaerobic conditions were experienced at Station 4 at 2330 hrs, some seven hrs after cessation of the rains. Evidence of the low oxygen plume was observed at station 6 between 1300 and 1500 hrs of the following day, with oxygen decreasing from 7.0 to 4.5 ppm. After traveling 12.6 km the plume was, unfortunately, not sufficiently oxygenated to meet the tolerance levels reported for most sports fish (10).

In contrast, oxygen levels below the S. Haven CSO decreased only slightly immediately after the rainstorm, although a release of untreated sewage was observed. Decreased perturbations are apparently suppressed by the increased discharge along this section of stream and more efficient aeration of effluent before release. Velocity of the contaminated plume ranged from 0.98 km/hr between Stations 3 and 4 and 1.33 km/hr between Stations 4 and 5. These calculations suggest a mean stream velocity of 34 cm/sec between stations 3 and 6, and are in general agreement with flow measurements taken during routine monthly outings.

During the August diurnal intermittent rains again resulted in even more prolonged anaerobic conditions downstream from the Valparaiso CSO. Station 2 was added as a control during this and subsequent outings and did not show significant perturbations associated with rainfall events.

The contrast between the bottom fauna of Station 2 and 3 further illustrates the impact of the Valparaiso CSO. Shannon Weaver Species Diversity Index, after Wilhm (14), above the outfall is nearly 9 fold greater than below it. The substrate underlying the riffle at Station 2 is largely composed of heterogenous concrete rubble. While Station 3 is also a riffle run, its bottom is composed of more uniform gravel in the center, while banks consist largely of deposited sludge. Although Station 2 should be expected to have higher diversity due to its greater flow rates and more heterogenous stoney substrate (4), this in itself probably does not account for the species diversity differences found. Both center and marginal areas of Station 3 are overwhelmingly dominated by the aquatic roundworm, *Tubifex*. This species is well known to be tolerant of organic pollution and its presence in large numbers is indicative of organic loading from sewage (3). In contrast, the bottom fauna of Station 2 is composed of a more balanced "clean" water community. For instance, *Baetis*, Heptagenniidae, Cordulidae, *Hydropsyche*, *Limnephilus*, *Stenemis*, Tipulidae and several pollution intolerant subfamilies of midges generally occur at this location.

Summary

Analyses of dissolved oxygen, specific conductance, pH, ammonia, nitrate, total phosphate, sulfate, chloride, sediment oxygen demand and organic content and biological species diversity during July, Aug., Sept. 1980 and Apr. 1981 of Salt Creek waters indicated that the Valparaiso Combined Sewage Outfall (CSO) was the major contributor to the degradation of the stream's water and sediment quality. There was less evidence of conspicuous negative impacts from South

Haven's CSO and most other point discharges into Salt Creek. The study revealed little evidence of organic and sediment loading from agricultural practices. Sand mining operations at the headwaters of Sager Creek above Sager Lake may significantly impact upstream ecosystems and potential salmon spawning areas. An undocumented sewage outfall between Horse Prairie Road and U.S. Highway 30 is strongly suspected although water quality data obtained at this point was inconsistent. Despite heavy pollution imposed on the stream by these point sources, the stream at base flow conditions appeared capable of limited recovery about 9.0 to 12.6 km downstream of the Valparaiso CSO. Unfortunately, the heaviest pollution took place at stream locations with the greatest salmon spawning potential. Diurnal analyses indicated that episodes of significant rainfall coincided with anaerobic conditions in areas downstream of the Valparaiso CSO and with depressed oxygen levels as far north as the State Highway 6 station. Under these water quality conditions, possible sediment loading from sand mining operations, and limited suitable spawning substrates, it is unlikely that Salt Creek's potential as a salmon fishery will improve.

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GEOLOGY AND GEOGRAPHY

Chairman: JOHN H. CLEVELAND
Department of Geography and Geology
Indiana State University, Terre Haute, Indiana 47809

Chairman-Elect: TERRY R. WEST
Department of Geosciences
Purdue University, West Lafayette, Indiana 47907

ABSTRACTS

Environmental and Engineering Geology of Hancock County, Indiana. G.W. CROSBY and T.R. WEST, Department of Geosciences, Purdue University, West Lafayette, Indiana 47907.——The purpose of the study is to provide information on environmental and engineering geology of earth materials in Hancock County for engineers and land use planners. Data are presented through maps, plates, cross-sections and tables providing for detailed interpretation in planning and design.

Figures depict topography, drainage development, watershed boundaries, current land use, soil associations, and ground-water availability. Twenty-six cross-sections using more than 200 water-well logs yield information on glacial geology, ground water, waste disposal, and aggregate availability. Plates depicting the county at a 1″ = 1 mile scale illustrate surficial geology, glacial drift thickness, and bedrock topography. Additional information on soil series is presented in table form. For the town of Greenfield, potential sanitary landfill sites are designated within a five-mile radius.

Proper application of supplied data enables engineers and planners to 1) locate areas with favorable geologic conditions relative to a proposed land use 2) eliminate unsuitable areas for such use 3) recognize areas with potential for aggregate supply and 4) avoid locations which could lead to environmental hazards. Despite the specific geologic information provided, an on-site investigation likely including a subsurface investigation, is needed prior to final site selection. This study was supported in part by a grant from the Indiana Academy of Science.

The Renault Formation (Mississippian) in Southwestern Owen County, Indiana. GEORGINA L. CUNDIFF, F.J. Reitz High School, Evansville, Indiana 47712 and PADMAJA D. MANERIKAR, Rich South High School, Richton Park, Illinois 60471.——Sedimentologic history of the Renault Formation in the subsurface of southwestern Owen County was interpreted by utilizing well samples, core chips, and geophysical logs from 65 wells. The Aux Vases Formation underlies the Renault Formation. A thin zone of shale separates up to 26 feet of the Renault into an upper and lower limestone.

The crinoidal wackestone and packstone of the lower Renault were deposited in a subtidal marine environment under moderate wave and current activity. The oolitic grainstones of the upper Renault formed in very shallow, highly agitated shoaling waters. Superadjacent rock, including fine-grained wackestone at the Renault top and the shale and siltstone of the overlying Bethel Formation, document an increase in water depth. The structure of the Renault top indicates a gradual southwestward dip with concentrated local folding related to differential compaction above Silurian reefs over 1000 feet below the Renault. The Renault

Formation thins to less than 15 feet in the general positions above the Silurian reefs. More than 145 feet of younger Mississippian rocks above the Renault are preserved in some locations, but a pre-Pennsylvanian stream valley was cut below the Renault in the eastern part of the study area.

Indianapolis: A New Urban Image. RAHIM KHAVIDI, Department of Geography, Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana 46202.——This paper takes a new look at Indianapolis and its immediate urban region in a geographic perspective focusing on its recent developmental planning policies and programs for growth and survival. First, the study briefly reviews the city's steady population and spatial growth from its first settlement (early 1820s) through the city's constant population growth (late 1960s). Second, the study overviews the population trends during the late 1960s which resulted in an net out-migration of 71,413 persons from the city to the adjacent suburban fringe, both within and outside of Marion County. The combined effect of population shift and out-migration caused a declining central-city tax base, abandoned housing, and the underutilization of urban services in the central city. Moreover, the growing urban sprawl created a high demand for urban services in the outlying areas throughout the 1970s.

Third, the study concentrates on the recent (1970s and after) urban and economic growth policies designed to halt the increasing net out-migration of residents from Indianapolis and to make the central city and downtown attractive for prospective residents and commercial establishments. This has been accomplished by various downtown and neighborhood revitalization projects, motivating public and private sources to reinvest in and improve existing urban services, facilities, and amenities as well as to expand the opportunities for new growth. In this regard, this paper will highlight many of the recent revitalization, economic, educational, and sport projects around downtown, as well as the rapidly expanding campus, of Indiana University-Purdue University at Indianapolis.

Determination of Pore Size Distribution in Soils and Rocks. C.W. LOVELL, School of Civil Engineering, Purdue University, West Lafayette, Indiana 47907.——All of the properties of soil and rock important to engineering use are influenced by the value of porosity and its distribution. It is now possible to make measurements of pore size distribution in a routine manner via the mercury intrusion technique. The mercury is pushed into the pores of an evacuated sample, with concurrent measurements of absolute pressure and volume intruded. The ordinary capillary equation allows the calculation of smallest pore diameter intruded at a given pressure. By successive increases of pressure on the mercury, smaller and smaller pores are intruded, and a cumulative curve of volume of pores vs. pore size is generated. The distribution may be converted to a differential form if this is helpful in interpreting and correlating the data.

A number of engineering properties of compacted soils and mineral aggregates have been correlated with their pore size distributions. The most prominent of these have been the water permeability and frost susceptibility of compacted soils, and the physical durability of shales and carbonate rocks. Examples are presented.

Aligned Kettles, Morgan County, Indiana. TIM MORRIS, Metropolitan School District of Martinsville, ROBERT C. HOWE Department of Geography and Geology, Indiana State University, Terre Haute, Indiana 47809 and NED K. BLEUER, Indiana Geological Survey, Bloomington, Indiana 47405.——Approximately three miles northeast of Martinsville, Indiana, in a region where the boundary of Wiscon-

sinan glaciation has a north-south orientation, about 18 topographic depressions which appear to be kettles exist within a one square mile area. These roughly circular pits, which average about 100 m in diameter, are filled to varying degrees by sediment and water. Peat has been extracted from one of them for a golf course. Several are dry at the surface but become lakes after heavy rains. Two of the basins are "double kettles". Of particular interest is the apparent alignment of the depressions mainly in a NE-SW direction but also in a NW-SW alignment of associated ridges. The NW-SW orientation parallels a NE-SW alignment of associated ridges. The NW-SE alignment appears to follow a pre-existing valley but may correspond to an ice marginal position. Thin and discontinuous Wisconsinan till occurs in that part of the area which has historically been mapped as underlain by pre-Wisconsinan till and/or outwash. Significantly, the area corresponds with the place where southwestward-moving Wisconsinan ice moved off high, impermeable siltstone bedrock onto the westward-thickening wedge of permeable, pre-Wisconsinan outwash.

Use of Chemical Surfactants in Disaggregating Argillaceous Rocks. SUZANNE PETERSON, Anoka High School, Anoka, Minnesota 55303, CHRISTOPHER G. MAPLES AND N. GARY LANE, Department of Geology, Indiana University, Bloomington, Indiana 47405.——Breaking down shales and mudstones to release small fossils or other embedded fragments, without damage to limy skeletons, has traditionally relied on either soaking in a petrochemical (kerosene, oleum spirits, and so on) or boiling them in a single chemical surfactant, Quaternary-O (Zingula, 1968). We here report on trial preparation of eight different argillaceous rocks, ranging from soft, marly mudstone (Bangor Formation, Alabama) to siliceous (Fort Payne Chert, Tennessee) and black carbonaceous (New Albany Shale, Indiana) shales. Five commercially available and three experimental surfactants were used, and diluted to a 0.01 molar solution. Each sample was boiled for four hours, the pH of each solution was determined, both before and after boiling, and the sample massed and sieved (20 and 250 mesh) after drying. Quaternary-O was used as a standard for comparison. The masses were used to calculate the percentage of disaggregation of the shale. Results varied widely from shale to shale and surfactant to surfactant, but Quaternary-O and Experimental Surfactant 2 worked best overall. Individually, however, each of the surfactants tested did as well or better than Quaternary-O on individual samples. For example, FC-760 did better on the New Albany Shale than Quaternary-O.

A Structurally and Stratigraphically Anomalous Area near Charlestown, Indiana. PETER A. WORCESTER and JAMES E. MCNULTY, Department of Geology, Hanover College, Hanover, Indiana 47243.——A small area about two miles east of Charlestown, Indiana, is anomalous both stratigraphically and structurally with respect to the rest of southeastern Indiana. The surrounding stratigraphy, typical of much of southeastern Indiana, consists of the Dillsboro, Saluda, Brassfield, Salamonie, Waldron, Louisville, and Jeffersonville Formations. Within the anomalous area, a breccia apparently replaces part of the Saluda, the Brassfield, and part of the Salamonie. Above the breccia the Laurel Member of the Salamonie Formation is atypically rich in chert.

Locally within the area, the Dillsboro and Saluda Formations are steeply inclined (as much as 30°) and are cut by small thrust faults. Although their relationship is not yet clear because outcrops of the breccia and especially the deformed beds are few, it is assumed that the breccia is related to the disturbed strata.

Four possible causes of the anomaly are being considered: 1) submarine sedi-

ment flows; 2) local tectonic adjustment during the development of the Cincinnati Arch; 3) meteorite impact; and 4) a combination of 1 and 2 or 3.

Roof Characteristics of the Springfield (No. V) Coal in Indiana

S. N. Ghose
AMAX Coal Company, Indianapolis, Indiana 46206

Introduction

In Indiana the Springfield (No. V) coal is the most persistent seam with the largest in situ reserve and greatest mineable potential. Approximately 95% of the remaining in situ 11.7 billion tons of Coal V estimated by the Indiana Geological Survey has to be extracted by underground mining methods. By virtue of its being in the median stratigraphic position among the major mineable seams, the bulk of the reserve occurs in the shallow underground mineable category, which is less than 600 feet deep.

In any method of underground coal extraction, the nature of the roof above the coal seam is one of the key factors in determining the economic mineability of the seam.

In the present study an attempt has been made to formulate the basic characteristics of the Coal V roof in Indiana and to define their variabilities on a regional scale. The specific details of the localized target areas for mining have not been discussed here primarily due to the lack of extensive data control and the uniqueness of each mine-block.

Geology of the Roof

The various lithologic units overlying a coal seam in a composite form are collectively called the roof of the seam. Geologically a roof can be divided into two categories—immediate or primary and main or secondary. Immediate roof can be defined as the lithostratigraphic unit which lies directly above the coal seam. The main roof which overlies the immediate roof usually is composed of multiple lithostratigraphic units. The total thickness or upper limit of the main roof can be defined either, by the direct influence of the uppermost lithostratigraphic unit can have on the stability of the roof system, or the lithostratigraphic unit used as the foundation for the roof support mechanism. In general, the immediate and the main roof constitute a heterogenous geological condition. The homogenous geologic condition in a roof system exists when the immediate roof is very thick and extends vertically beyond the zone of influence. In the heterogenous roof system the two categories are not always complimentary of each other.

To be consistent for the purpose of generalization, a fixed 10 feet zone above the coal seam has been adopted as an index of evaluation. Several hundreds of these '10 feet zones' above the coal seam from representative geographical distribution have been utilized to define the litho and chronostratigraphic variabilities.

The immediate roof of Coal V in Indiana can be defined as a member of the Petersburg Formation which shows lithologic variations geographically over the Indiana coalfields. Except for a few localized areas where the Alum Cave Limestone overlies Coal V, the immediate roof is considered to be time parallel or formed contemporaneously. The total range of changes in the immediate roof is probably caused by changes in the depositional environment. As is shown in Figure 1, the immediate Coal V roof in Indiana is predominantly black shale. Although the black shale for mapping convenience has been characterized as a

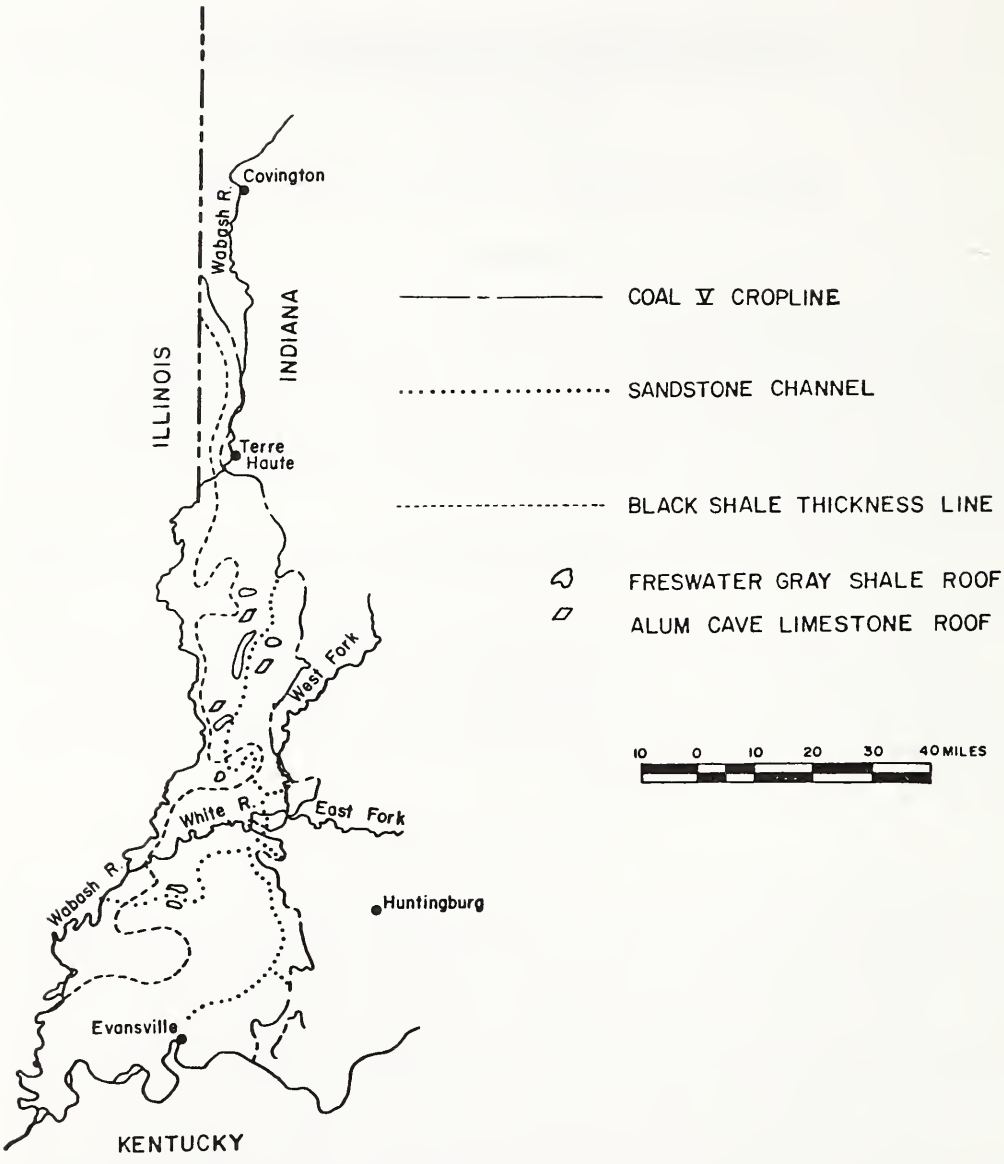

FIGURE 1. *Geographical distribution of the immediate roof above Coal V in Indiana. (Black shale roof is present in the entire area, except gray shale and limestone locations.)*

uniform member in the entire coalfield, close field examinations reveal that there are few distinctions in properties which can influence the mineability of Coal V. Also, as shown in Figure 1, a generalized thickness line delineating the "more than 3 feet and less than 3 feet" of black shale areas is presented. The characteristics which are prominent in these two divisions are tabulated below.

	+3 feet	3 feet
1.	typically massive	typically laminated
2.	comparatively harder (bulk hardness)	brittle and breaks along the horizontal planes
3.	silty	more clayey
4.	typical area - Vermillion and northern Sullivan counties	Warrick County

The laminated type of black shale probably was deposited in the subenvironment where the water was shallower and quieter.

The second variety of immediate roof which is present geographically in very localized areas is composed of fresh water gray shale. This is associated with the contemporaneous sandstone channel in Coal V, as shown in Figure 1. Typically, the gray shale is massive, moderately soft, clayey and becomes sandy towards the channel. The contact with black shale is usually sharp and distinct. Unlike the extensive gray shale deposits in southern Illinois, the thickness of gray shale in Indiana is typically restricted except where the gradual sandy phase overlaps.

In isolated areas of very limited extent, the immediate roof is composed of argillaceous limestone—Alum Cave. It is possible that in these areas due to adverse depositional conditions, the black shale was very thin or was not deposited and later erosion removed the remainder.

Typically the main roof consists of the lithologic units which are geologically younger than the immediate roof and the coal seam. Each lithologic unit in the main roof is primarily a function of time and the lithologic units are progressively younger upward. In Figure 2, the representative number of lithologic

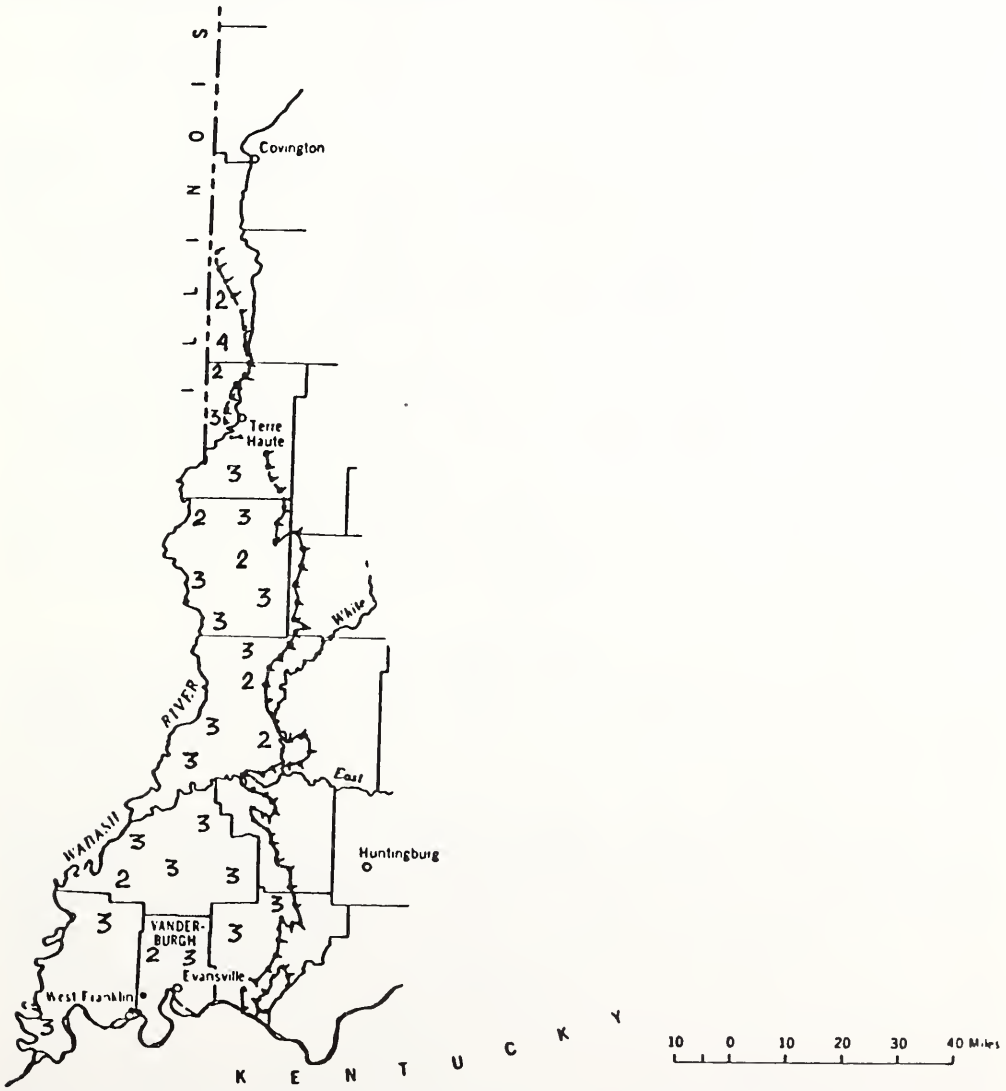

FIGURE 2. *Number of lithologic units present in the first 10 feet above Coal V.*

units in the main roof for different geographical locations is shown. These represent best estimates of averages (based upon the available date) for any given area. These lithologic units are either members of the Petersburg or the Dugger Formation. Based on the number of lithologic units present in the remainder of the interval, a general approximation about the stability of the roof can be obtained (typically for Indiana coal fields). It is assumed here that each lithologic unit is adding one plane of weakness. Thus, the stability of the Coal V roof is inversely proportional to the number of lithologic units present in the main roof, or the stability of the Coal V roof = K x number of lithologic units present in the main roof. The value of K, constant of proportionality will vary from place to place depending upon the local geology, nature of lithologic units and the past history of mining. In this approximation, the orientation of excavation either parallel or perpendicular to the bedding planes has not been considered.

In Figure 3, different combinations of qualitative variations have been illustrated. Each combination gives an indication of the stability of the roof relative to the lithologic characteristics. Laterally, the lithologic variations for the members of the main roof are very abrupt and inconsistent. It is probable that during late Petersburg and early Dugger time, the depositional system was very active, variable and restricted.

Structure and Physical Properties

In the preceding section, the range of lithologic variations in the immediate and the main roof of Coal V in Indiana was described. A generalized geographical distribution of the rock types in these two categories of roof has also been presented. The physical characteristics and the sedimentary structures associated with these rock types will be briefly discussed here.

Black Shale: Typically, the black shale in Indiana is very dark to moderately dark, moderately hard and contains varying amounts of organic materials.

The laminated type of black shale which is rather brittle and breaks along horizontal planes is relatively smooth and even. The cracks are rather prominent and relatively less water sensitive than the massive type of black shale.

The massive type normally shows irregular cracks, lenses of silty materials and contains slightly coarser sediments. It is relatively more water sensitive than the laminated type. Near the contemporaneous sandstone channel in places, the normal sequence of lamination is disturbed and lenses of silty materials become more prominent.

The laminated black shale frequently parts unevenly along the horizontal planes, activated by the presence of fluids in exposed conditions. The massive variety is moderately competent except where it contains lenses of silty materials, irregular bandings, etc. The black shale roof type in Indiana can be considered more competent than the fresh water gray shale.Gray shale (fresh water): Light to moderately dark, massive, invariably silty, porous, moderately hard to soft and breaks unevenly. The gray shale in Indiana is typically inhomogenous, inconsistent and not very thick. Numerous lenses of fine grained sand are typically present. Close to the channel the gray shale shows distortion in restricted areas. Irregular, interconnecting cracks are common. Because it is typically water sensitive and disintegrates easily, the fresh water gray shale commonly exhibits poor roof conditions with a very limited holding capacity.

Alum Cave Limestone: It is a light colored, argillaceous, 4 to 5 feet thick, moderately hard to hard limestone with an uneven fracture pattern. Typically, it occurs in two benches: The lower bench is thinner, compact and even. The two benches

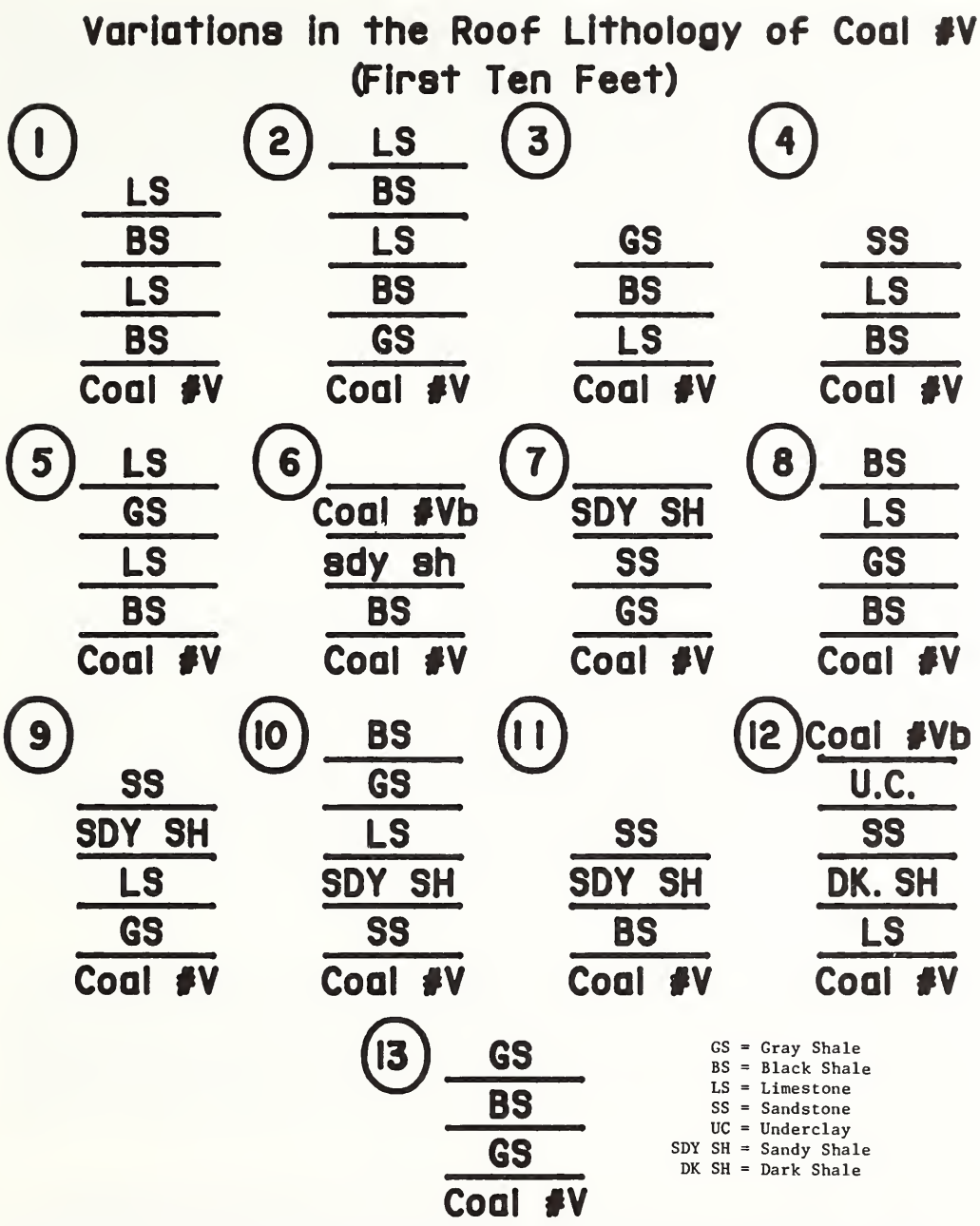

FIGURE 3. *Various lithologic combinations present in the first ten feet above Coal V in Indiana.*

are typically separated by black or marine gray shale. The upper bench is normally shaley, less compact and uneven. Generally the fractures are irregular, intersecting and unevenly distributed, and the shaley areas are moderately water sensitive. If the lower bench is reasonably thick (> 2 ft.), and consistent, the Alum Cave affords good roof conditions in underground mining.

Discussion

From the data presented in the preceding sections, it is evident that local roof variations are locally more pronounced compared to variation on a regional scale in Indiana. Hence, based on the regional trends we can devise a scheme

of assessment which can be applied to determine the competency of a roof system in a given localized area. This scheme may be helpful as a first approximation in the early stages of exploration where very little detailed information is available.

This is a semiquantitative scheme based on the comparative weight factors on a scale of one to ten, assigned qualitatively without any bias. Even with some degree of bias the gross result will remain unaffected. The various parameters in the assessment are given below.

1.) Nature of the immediate roof—the most favorable (realistic) situation in Indiana can be at least three feet thick, Alum Cave Limestone without any inclusion of shaley materials (compressive strength > 10,000 PSI). A relative scale of competency based on the megascopic characters for assigning weights, in decreasing order of preference is as follows:

Lithologic units:	limestone (> 3 ft.)	finegrained hard sandstone	black shale	compact sandy shale	soft fresh water gray shale
Weight :	10	8	6	4	2

Homogeneity and thickness are two variables which should also be considered while assigning weights in this category. Thick and homogenous lithologic units are favorable unless the units are composed of incompetent soft materials.

2.) Depth is also an important parameter. Because, the same marginally competent materials at shallower depth may not be competent at a substantially greater depth. Also, groundwater seepage, structural features and discontinuities would increase the susceptibility to failure at greater depths. An example of a simplified scale of weights based on depth is given below:

Depth (ft.) :	200	400	600	800	1,000
Weight :	10	8	6	4	2

3.) Number of unit beds present in the main roof is an important parameter to be weighted. In general, the fewer number of units in the main roof, the more stable the system is for underground mining. In the case of the multi layered main roof situation, the effect of weakness is cumulative to the process of deformation. Weights can be assigned in the inverse order to the number of units.

Number of lithologic units in the main roof :	10	9	8	7	6	5	4	3	2	1
Weight :	1	2	3	4	5	6	7	8	9	10

Geologically, if the changes are not gradational, by addition of one more units in the system, the homogeneity is decreased to some extent and thus the probability of failure increases.

4.) Lithologic nature of the units present in the main roof is also important and should be weighted accordingly. For example, if in a main roof system the lithologic units are alternating fresh water—soft gray shale and coarse grained loosely cemented sandstone, then this category should be given lower weights. The weights can be assigned as in the immediate roof (parameter 1).

5.) Position of the relatively incompetent bed and the arrangement of the lithologic units in order of competency in the main roof system is also an important parameter.

There can be three orders of arrangement—vertically increasing order of com-

petency, vertically decreasing order of competency and irregular order. Each one has its own merits and demerits and should be evaluated with respect to the immediate roof and the basal unit of the main roof. If the basal unit is thick, then a vertically decreasing order of competency can withstand the overlying load, but if the basal unit is thin, then a vertically increasing order of competency is needed for support. In the case of an irregular order, the position of the most incompetent bed in the system is important. In Figure 3, 13 lithologic combinations are shown and weights can be assigned in the following manner.

Combination :	1	2	3	4	5	6	7	8	9	10	11	12	13
Weight :	8	6	8	8	6	4	4	6	4	6	6	6	2

5.) The density of cracks, joints and fractures present in the unit area should also be considered. Length and orientation of the cracks are critical. Under pressure, the cracks tend to connect and create roof falls. Also the presence of fluid in these cracks increases the chances of failure. If under pressure, the cracks connect with each other, the bulk permeability of the rock is increased which reduces the in situ compressive strength. Hence, if the cracks are close together and oriented at angles to one another where two sets of cracks intersect, these should be assigned lower weights.

6.) Water sensitivity is also an important parameter which is detrimental to the roof stability. Typically, the clayey shale and loosely cemented sandstone are the most susceptible to water. The expendable clay minerals like montmorillonite, if present, will swell and reduce the compressive strength. The weights should be assigned, based on the results of field water sensitivity test.

Based on the above parameters, a semiquantitative estimate of the roof stability in a given area can be determined which bears directly on whether the reserve has potential for detailed analysis.

Conclusion

Interpretation of geological variables to determine the stability of roof, unless quantified, does not represent the sensitivity of correlation. Qualitative judgements do not properly verify the factor of risk and the uncertainties associated with the projected inferences. The present study needs refinements in order to be effective at the final stages of mine planning.

Acknowledgments

This study represents a research done under the guidance of Dr. H. H. Murray of Indiana University. I am thankful to Mr. D. A. Darko for giving permission to publish this paper. The assistance in critically reviewing the manuscript by Dr. C. E. Wier and Mr. T. R. McCarthy is gratefully acknowledged.

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HISTORY OF SCIENCE

Chairman: GENE KRITSKY
Department of Biology
Tri-State University, Angola, Indiana 46703

Chairman-Elect: GENE KRITSKY
Department of Biology
Tri-State University, Angola, Indiana 46703

ABSTRACT

Charles Darwin and Bees. GENE KRITSKY, Department of Biology, Tri-State University, Angola, Indiana 46703.—— Charles Darwin's interest in insects extends back to his boyhood days, when he collected beetles as a hobby. Although he did not write a book solely dedicated to the insects, Darwin did use numerous examples of insect behavior, genetics, evolution, and morphology in his theories of evolution and pangenesis.

With the publication of an index to Charles Darwin's insect references (Kritsky 1981), it is possible to examine all of Darwin's writings including his letters, to determine his interest in bees. Darwin was interested in honey bees and bumble bees. His interest was primarily in the area of cell or comb construction, and genetic variation.

Darwin used comb construction as an example to show that instinct evolved. By looking at examples in the bee family, Darwin was able to show a gradation in behavior in comb construction, from simple cells, to the complex colonies of the honey bees.

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A Brief History of Biology at Indiana University

Frank N. Young
Department of Biology
Indiana University, Bloomington, Indiana 47405

Indiana University is the oldest state university west of the Allegheny Mountains.[1] It was established by law as the Indiana Seminary by the legislature in 1820, became Indiana College in 1828, and Indiana University in 1838. The university became coeducational in 1867. Biology Hall now Swain East was finished in 1910. Jordan Hall of Biology was occupied in 1955.

The first self-conscious courses in biology were offered in Bloomington in the academic year 1853-1854, but even as early as 1828 *John H. Harney* was appointed to teach mathematics and "such of the natural sciences as were considered of sufficient importance to engage the attention of aspiring youth." The faculty at that time consisted of three professors. We do not really know what was taught in the early days, but we may surmise from the textbooks used that there was a lot of anatomy, a little embryology and physiology, some biogeography, and much systematics. Certainly not the smorgasbord of modern biology from which students may choose today.

The early socialistic experiments of *Robert Owen* at New Harmony, Indiana, produced an important feedback with the addition of Robert Owen's son Richard to the I. U. faculty in 1864. *Richard Owen* was a remarkable man. He graduated from the Nasvhille Medical College in 1858, but apparently never practiced medicine. He commanded the 60th Indiana Volunteers during the Civil War, and must have been an exemplary officer because in the entry to the I. U. Memorial Union is a tribute to him provided by contribution from Confederate prisoners who were confined in the prison camp which he commanded. There is no group of people under the sun less likely to thus honor a man without due cause.

Richard Owen's chief interests were in geology rather than biology as such, but to judge from the records, a great deal of zoology and botany were included in his teaching. Even the then new ideas about organic evolution were not neglected although they were probably not as central a theme as they are today. Richard Owen, in his teaching, often mentioned the names and work of such greats as Cuvier, Linnaeus, Darwin, and Huxley. By the 1869-70 academic year, zoology and botany appeared as separate courses among the natural sciences.

Richard Owen's tenure at I. U. terminated in 1879. He was followed by a more remarkable man, *David Starr Jordan*, after whom Jordan Hall, Jordan Avenue, and the Jordan River were named. The manner of Jordan's appointment is somewhat curious. He was then teaching at Butler University in Indianapolis and came to Bloomington to speak on behalf of a friend who sought an appointment at I. U. The trustees unanimously voted to offer the professorship of natural sciences to Jordan. Later in 1884 a somewhat similar incident resulted in Jordan's accedence as president of the university. He had been asked to recommend someone else and again was unanimously selected. *David Starr Jordan* may have been the last person anyone would want to send to speak for him, but there is absolutely no imputation that he was not a man of the highest moral and ethical character.

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1. I. U. is actually the oldest established by a state legislature. Michigan claims precedence as a church-related college before statehood.

Jordan did not assume his duties as Professor of Natural Sciences immediately. As *W. J. Moenkhaus* commented in the *Indiana Student*, although he was appointed in 1879 he was granted a year off to go fishing. Actually he was collecting fish in the Pacific Northwest under the auspices of the U. S. Fish Commission. During his years at I. U. he continued his research on fish, and aquatic biology has remained one of the strong areas in I. U. biology down to the present.

Following the leadership of Jordan, biology, and later the whole curriculum of the university, was changed. I think we can say that it was modernized because we still maintain many of the central themes of Jordan's reforms. One of these is the free elective system under which a student has "the right to choose in accordance with his own powers and tastes." Of course we have had to provide some guidelines for this procedure to protect the guileless ones. In 1886-87 the university curriculum was reorganized on the major subject and departmental basis.

In the summer of 1883 an event occurred which greatly modified the future of I. U. This was the burning of the then new Science Building near what is now College and West Second Street (now designated "Seminary Square"). The loss of the Science Building with all the collections and library spurred a move on the part of the university to its present site on what was then called Dunn's Woods. Owen and Wylie Halls, named after *Richard Owen* and *Andrew Wylie* (the first president), were the first buildings erected in the central quadrangle. A wooden structure still standing may have been the first building on the new campus to have classes held in it.

An interesting little sidelight on the library was that after a new library was organized, the loss due to the 1883 fire turned out not to have been as great as it might have been. A number of professors had all the important books in their fields in their offices or at home, and after the new library was available books which were thought lost began to be returned.

In 1884 Jordan became president and was free to use his charismatic personality on the trustees and faculty. He brought about a veritable revolution in biology and in the university as a whole. Despite his administrative duties, he continued his own research. Years later, *Barton W. Everman*, the co-author with Jordan of massive works on the fishes of North America, paid this homage to Jordan at a meeting of the Indiana Academy of Science which Jordan helped to found in 1885:

> "The greatest impetus ever given to zoological research and investigation in Indiana occurred when *David Starr Jordan* came to Indiana in 1874 as a teacher of natural history.The twelve years spent by Dr. Jordan at Indiana University were among the most productive of his life, not only in relation to zoological science in general, but to zoology in Indiana in particular. The influence upon the state was epoch-making. The effect of training so many of its young men and women in methods of science and sending them out over the state and beyond its borders imbued with the spirit of the real naturalist who seeks truth, who sees things as they are, and who knows animals when he meets them in the open, cannot be overestimated. Many and varied were the problems in zoological science that these young men and women investigated, studied, and attempted to solve. They were by no means confined to the fauna of Indiana. In Ichthyology their field was world wide. It is true, however, that the richness of the Indiana fauna appealed to many of these young naturalists, and the zoological literature has been greatly enriched by their contributions."

Jordan's influences were not confined to zoology. The teaching in botany, geology, chemistry, and other areas was affected by his ideas and reforms. When he left in 1891 to become president of Stanford University, he was succeeded as head of the Zoology Department by *Carl H. Eigenmann*, who was succeeded by *Fernandus Payne*, who was succeeded by *Theodore W. Torrey*. These three men bridge the gap between the 19th century and the present.

I cannot leave *David Starr Jordan*, however, without a few comments. He made many important contributions to the educational program at I. U. including the summer session and extension education. Yet in his voluminous correspondence he seldom mentioned I. U., but dealt extensively with the importance of his own research. He was an elitist in education although it must be admitted an elitist of intellect not wealth or political power. He left precipitously in 1891, apparently with little regret although he made a moving address to the faculty and students on his departure. Rumor has it that *Leland Stanford* offered him a bonus for each top-notch faculty member he took with him. His bounty hunting cost I. U. 12 of the best of its then 29 faculty members. Even more heinously, he also took with him 39 of the best students including the cream of the football team.

The Botany Department (later Plant Sciences), established by Jordan in 1885, has had an equally distinguished series of leaders. *John Merle Coulte* (1851-1928) was president of I. U. from 1891 to 1893 and also taught a course in botany. Bacteriology was first offered separately in 1896, and a full curriculum was introduced in 1940 by *Leland McClung*. This remains as the core of Microbiology. This field too has had active and able leaders beginning with McClung, *Howard Gest*, *Dean Fraxer*, and *Eugene D. Weinberg*. Recently the three former departments were united under the Department of Biology with *John Preer* as first chairman.

With *Carl H. Eigenmann* we enter the modern period of biology at Indiana University. Eigenmann's interests were primarily in fishes, but he and his students undertook studies of many aspects of evolutionary biology. He established in 1895 one of the first field stations in the United States at Turkey Lake (now called Lake Wawasee). The original objectives were to study the fauna, flora, and environmental factors in detail to see what correlation they had with the variation of organisms. Eigenmann became Professor of Zoology in 1891 and in 1894 Dean of the Graduate School. His influence was considerable. He assembled about him a faculty of able people, and at the time of his death in 1927 the biology faculty included *Will Scott*, *Fernandus Payne*, *A. C. Kinsey*, *David M. Mottier*, *Paul Weatherwax*, *Frank M. Andrews*, and *James Van Hook*. By this time physiology and anatomy had more or less drifted away following a separate course correlated with the medical program.

When Eigenmann died in 1927, the reins of Zoology and the Graduate School passed to *Fernandus Payne*. During his tenure, biology at I. U. reached one of its highest points. Distinguished faculty were added to all the science departments as well as biology. These included among others in zoology protozoologist *Tracy M. Sonneborn*; Nobel Laureate geneticist *H. J. Muller*; endocrinologists *Robert L. Kroc* and *William R. Breneman*; limnologists *William Ricker*, *Louis A. Krumholz*, and *Shelby Gerking*; ecologist *Lamont C. Cole*; biochemist *W. J. van Wagtendonk*, and embryologist *Theodore W. Torrey*. Botany added geneticist *Ralph E. Cleland*, biosystematist *Charles B. Heiser*, physiological ecologist *Stanley Cain*, mycologist *Harold J. Brody*, algologist Richard C. Starr, and physiologist *Charles W. Hagen*. Bacteriology was established as a separate department in 1946 under *Leland McClung* and added among others *Irwin C. Gunsalus* and *Salvador E. Luria* (later Nobel Laureate).

With the coming of the 20th Century the directions of biological research began to change. The rediscovery of Mendel's work was one important event. In part this discovery had a retarding effect on the development of biology. The "billiard ball" theory of genetic inheritance died hard. Today with discoveries in many organisms of multiple genes, duplicate genes, and even "jumping" genes this theory seems riduculous. It had, however, great appeal to some. It restored some of the stability which seemed to have been lost with the introduction of the concept of chance mutation acted upon by natural selection and the inception of the relativistic concept of the universe.

One of the greatest appeals of Darwin's proof of the fact of organic evolution was the neatness with which it fit the already established classification of animals and plants which had been worked out without any such process in mind. The new genetic theory also, eventually, proved to explain and to correlate with facts already discovered especially in the field of cytology. The "dances" of the chromosomes in mitosis and meiosis made perfect sense when correlated with Mendel's genetic ratios.

Cary Joyce (1949) says that the role of the novelist is to help break through the crusts which form on men's minds. One of the I. U. botanists had observed the double fertilization in maize before the end of the 19th Century, but was so sure that he knew the real truth that he threw away the slides as "abnormal" thus losing the opportunity of first reporting a most important phenonmenon. Fortunately, all of the I. U. biologists did not have such encrusted minds. *Fernandus Payne* soon began introducing the new science of genetics despite opposition from some of the "old guard."

With the election of *Herman B Wells* as president in 1938 and with the aid of new state retirement laws much of the "dead wood" was chopped out of the faculty as humanely as possible. Linked with a concerted effort to attract teachers and researchers of distinction and the phenomenal growth of the University after World War II this produced a new revolution in biology and the university as a whole.

The role of *H. B Wells* in the development of biology at I. U. cannot be denied. He was dedicated to excellence in every area of university life, and dedicated himself to achieving it. Under his leadership I. U. grew from a small, provincial school into an internationally recognized center of learning and research. His support toward efforts to obtain excellence was unfailing. His defense of the academic freedom of his faculty was always positive. Without this positive support work such as that done by *A. C. Kinsey* would have been impossible.

The influence of *Fernandus Payne* as Dean of the Graduate School as well as chairman of the Zoology Department extended far beyond biology in the reorganization and growth of the University. He came to I. U. in 1902, a simple Indiana farm boy backed with a tradition of absolute honesty and hard work. He evolved into a master cytologist and an administrator of great ability. After taking the Ph.D. degree at Columbia University, where he worked with *E. B. Wilson* and *T. H. Morgan*, he returned to I. U. in 1909. He was chairman of Zoology from 1927 until 1948 when he was succeeded by *T. W. Torrey*.

Payne was a courteous, very formal man. In his teaching he made his students learn. At the dedication of Jordan Hall in 1955, one of his former students, then chairman of zoology in an Indiana college, remarked to me, "Popsy Payne's courses were tough. I hated them when I took them, but I later realized I learned more in them than in any others I took at I. U."

In research, Payne worked mainly in the field of classical cytology but before leaving I. U. for Columbia he had worked with *Drosophila melanogaster* and his

paper "Sixty-nine Generations in the Dark" is a classical disproof of Lamarkian principles. He worked on the structure of the insect egg and widely on the chromosome structure of insects. He was among the first to recognize the involvement of the mitochondria with the nuclear material during spermatogenesis. He became Professor Emeritus in 1951, but continued working on spermatogenesis in *Gelastocoris* and other insects, taught himself to use the electron microscope which he recognized as creating a new horizon in cytology, and completed an intensive study of the cytology of the endocrine glands in aging. As a student he took *Drosophila melanogaster* from Bloomington to Columbia, and although he never acknowledged the source *T. H. Morgan* soon adopted it as an experimental animal. He was working on a final paper only a short time before his death. (See also Meserve, 1981, p. 5.)

One of the innovations of Jordan, Eigenmann, and *Fernandus Payne* was the attempt to involve all students in biology in basic research. Under Jordan many undergraduate students already had one or more research publications to their credit before graduation. The policy was not publish or perish but publish with pleasure. Many of those students never forgot that lesson and went on to distinguished careers in biology and other fields. With over 30,000 students on the I. U. Bloomington campus today we have had to modify this policy, but we still attempt to involve as many undergraduates as possible in research either during the regular semesters or the summer sessions.

Another concept stemming from long ago is that in graduate studies a student at I. U. must work with one of the faculty members competent to direct him and preferably directly in the area of that faculty member's interest.

The following notes on some of the biologists who have worked at Indiana University may be of interest to students. I have tried to include some analysis of their biological research as well as their service to Indiana University as teachers or administrators. T. W. Torrey's unpublished supplement to his 1949 paper should be consulted for details of several programs and individuals, especially for details of the sex research program initiated by *A. C. Kinsey*.

Research and Teaching at Indiana University
Anatomy and Physiology

The department of Anatomy and Physiology was separated from Zoology in 1904 after the establishment of the medical school. *W. J. Monenkhaus*, a student of Eigenmann's, was the first chairman. Later, anatomy and physiology were placed in separate departments and still later recombined. The development of the teaching and research programs in this area was largely tied to the medical program. Important research continues in these areas both at Bloomington and in Indianapolis, but much of its history is more closely related to medicine than biology as considered here.

However, I. U. physiologists and anatomists also have contributed basic studies in their areas. Currently *Roderick A. Suthers* is doing important studies on acoustic behavior in bats, and *Henry D. Prang* is studying the basic physiology and behavior of sea turtles. *Robert W. Bullard* did important work on comparative physiology of vertebrates at I. U. from 1956 until his untimely death in 1971. *Sid Robinson* did research on the physiology of exercise from 1930 until after his retirement, and *Bruce Dill*, after retiring at Harvard University, also worked here. Workers in other divisions of the University have also made extended studies in the physiology of exercise; notably *John M. Cooper* in the School of Health, Physical Education, and Recreation, and *James E. Counsilman* in the same school.

Aquatic Biology

With the interests of *David Starr Jordan* and *Carl Eigenmann* centered in fishes, aquatic biology had an early start at I. U. Eigenmann made pioneering studies on the blind fish of the cave system in what is now Spring Mill State Park. *A. M. Banta*, later at Brown University, did research as an undergraduate on the fauna of Mayfield's Cave just west of Bloomington, and other students made studies of aquatic habitats as well as fishes.

The development of a modern program in aquatic biology, however, began with the appointment of *Will Scott* as instructor in 1908. He received the A. B. and A. M. degrees from I. U. that same year and the Ph. D. in 1911. For many years he taught the large beginning course in zoology and directed students in work on lake and stream morphometry and sedimentation as well as in more directly biological studies. His primary contribution was as a teacher and as director of the I. U. Biological Station at Winona Lake from 1920 until his death in 1937.

The I. U. Biological Station had by this time become the principal site at which summer courses in biology were offered by I. U. Even such courses as comparative anatomy, histology, microtechnique, and embryology were offered there rather than on the I. U. campus. These courses were not moved to Bloomington until after Will Scott's death. The field oriented courses offered at Winona Lake inspired a number of later eminent biologists not only in aquatic biology but even in other disciplines.

As a man, *Will Scott* was a paragon. To quote *T. W. Torrey* (1949):

> "*Will Scott* was widely informed and that knowledge accrued to the benefit of the student. I have heard Doctor Breneman (*W. R. Breneman*) say that he learned more general biology in Scott's course in limnology at Winona Lake than from any combination of other sources. From personal experience I can attest that *Will Scott* was one of the kindest, most generous and thoughtful men I have ever known. Many instances of his help and kindly criticism come to mind. As a cub instructor I needed that help and he gave it unselfishly. In the same way he gave time and energy toward the betterment of the community. Science and humanity could use more men like *Will Scott.*"

The lacuna in aquatic biology was filled in 1939 by the appointment of *William E. Ricker* as assistant professor and director of the lake station. Ricker revived the research program at the station and at I. U. *Louis A. Krumholz* (1909-80) was added to the staff in 1945 and *Shelby Gerking* in the following spring under an agreement with the Indiana Department of Conservation. Ricker, Krumholz, and Gerking developed a strong program of lake and stream studies throughout Indiana. Ricker is widely known for his studies of fish biology, productivity, the dynamics of fish populations, and the application of mathematics to aquatic studies as well as his taxonomic studies on stone flies. He left I. U. in 1950 to return to his native Canada where he has since occupied important positions in fisheries research and conservation, and was elected to the Canadian National Academy of Science.

Ricker was succeeded in 1950 by *David G. Frey* who, together with Gerking, continued the program in aquatic biology. Frey's interest in the chydorid crustacea has extended the work of himself and his students far beyond the boundaries of Indiana—from Alaska and Southhampton Island to the tropics and into Europe and Asia. Frey's research on crustacea, the history of lakes, and other aspects

of limnology have become basic in several areas of limnology. (See also Eoyang, 1979, pp. 19-21.)

The Indiana University Biological Station and the Indiana Lake and Stream Survey

The Biological Station established at Turkey Lake in 1895 by Eigenmann flourished in its early days. As noted previously it was not until 1936 that the teaching of many summer courses was moved back to Bloomington. In 1899 the station was moved to Winona Lake, and in 1964 a new and modernized laboratory building was erected at Crooked Lake near Columbia City, Indiana. *Shelby Gerking* was instrumental in developing this new facility. The station continued as a center for research by faculty and students into the 1970s, but continuing changes in research interests led to its administrative transfer to the campus of Indiana University-Purdue University at Fort Wayne in 1980.

The Water Resources Center

The completion of the large Lake Monroe reservoir just south of Bloomington about 1967 brought various concerns about aquatic resources to focus. The Water Resources Research Center was established at I. U. and it was envisioned that the Crooked Lake Station, a new station for which land was acquired on Lake Monroe, and the Research Center would all be under the direction of a single director. The distinguished British limnologist *H. B. N. Hynes* accepted this post, but before arriving at I. U. had second thoughts and remained in Canada. The Research Center now directed by *Robert V. Ruhe* remains under the direction of the Geology Department, but the Lake Monroe station was never developed. (See also Meserve, 1981, pp. 12-16.)

Botany

Botany and Geology were separated from Zoology in 1885. *John C. Branner*, later a distinguished geologist at Stanford University, was the first in charge of both geology and botany. When he left with Jordan for Stanford in 1891, *John Merle Coulter* became professor of botany and president of the university. Coulter's research was primarily in the morphology of angiosperms and gymnosperms. He later published a number of botanical textbooks, one of which included the then new field of plant ecology (1911, with *C. R. Barnes* and *H. C. Cowles*).

David M(yers) Mottier (1864-1940) received his doctorate in Bonn in 1897. He had joined the staff at I. U. in 1891 as instructor and on his return from Germany remained with the department as associate professor, professor, and head until 1938. His research was primarily cytological, but he did some important work on the effects of centrifugal forces on reproductive cells and on the nature of plastids.

Associated with Mottier was *James M. Vanhook* (1870-1935). He was a student at I. U. and on the botany faculty from 1907-35. His work was primarily with plant pathology and mycology. *Frank M. Andrews* (1872-1940) was also associated with the department from his student days until his death. He taught and did research in physiological botany.

Paul Weatherwax (1888-1976) began his teaching career in a one-room schoolhouse near Worthington, Indiana, in 1907. He began college courses at DePauw University in 1909, and continued at Wabash College and I. U. His college training was interrupted several times by the necessity of returning to teaching in order to obtain funds, but he received the Ph.D. degree from I. U.

in 1918. After a brief period of teaching at the University of Georgia he returned to I. U. in 1921 and remained here until his retirement in 1959. He did not, however, stop teaching, and he taught at Franklin and Hanover Colleges until 1966. His teaching career thus spanned nearly 60 years.

His principal research interest was in the morphology of grasses particularly maize or Indian corn. His early interpretation of the structure of the corn plant, especially its flowers, was basic for the understanding of the origin of maize. His *Indian Corn in Old America* published in 1954 summarizes much of his research on maize. He was also an expert draftsman, and among other works illustrated C. C. Deam's *Grasses of Indiana* (1929).

Paul Weatherwax' principal contribution to Indiana University was through his teaching, but he was interested in every aspect of university life and contributed largely through his service on many committees. One of his last papers, *The Woodland Campus of Indiana University* (1966), is still a valuable guide for faculty, students, and visitors. (See also Meserve, 1981, p. 4.)

Ralph E. Cleland (1892-1972) came to I. U. as chairman of the Botany Department in 1938 from Goucher. He became dean of the Graduate School in 1950. He was an outstanding administrator, teacher, and research worker.

> "His position in the history of science is secure, and today he stands as the foremost student of *Oenothera* (the evening primrose). As a young scientist, he made the critical observations on the behavior and structure of the chromosomes that led to an understanding of the unorthodox genetic mechanism in *Oenothera*. This was followed over the years by a series of beautifully executed investigations on the evolutionary history of the genus. He remained active in research to the last, and his book *Oenothera, Cytogenetics and Evolution*, summarizing his life time work and that of others, was sent to press the day before his death." (Hagen *et. al.*, 1972)

He died at his desk in Jordan Hall June 11, 1972.

Herbarium of Indiana University

The herbarium of Indiana University is presently housed in Jordan Hall. It now contains over 100,000 accessioned sheets of specimens of vascular plants. Of these some 73,000 came from the collection of *Charles C. Deam* and are mainly the voucher specimens for his "Flora of Indiana" (1940), and other books.

The herbarium began bravely in 1892 under the direction of *Jonh M. Coulter*, but two years later on Coulter's move to Lake Forest College the herbarium went with him cases and all. The collection was begun again in 1895. Various students and faculty members contributed to the collection so that by 1931 it consisted of nearly 10,000 sheets. *Martha Springer* was instrumental in initiating an extensive program of exchanges from 1944 to 1947, when *C. B. Heiser, Jr.* became the curator.

Besides the Deam material the herbarium also houses part of an extensive collection of oaks made by *A. C. Kinsey* and important material from the studies of Heiser and his students who have worked extensively on wild and cultivated plants.

Drosophila Genetics

H(ermann) J(oseph) Muller (1894-1967) came to I. U. in 1945. He had previously taught and done research at the University of Texas, in Germany, the Soviet

Union, Scotland, and at Amherst College. He received the Ph.D. degree from Columbia University in 1916 under the direction of *T. H. Morgan*. His research interests were primarily with the fruitfly, *Drosophila melanogaster*. He made basic contributions in the area of linkage of genes, crossing over, multiple factor inheritance, the theory of the gene, gene mutations, chromosomal changes and their artificial production, the biological effects of radiation, the properties of chromosome parts, and in the broad area of evolutionary theory, speciation, and human genetics. His work on the effects of radiation resulted in his being awarded the Nobel Prize in 1946.

One of Muller's special talents was in the production of genetic strains of *Drosophila* with which various hypotheses could be tested. His designs, written mainly on small cards, showing the way into, through, and out of these crosses were often as intricate as those of any medieval necromancer. A significant difference was that Jo's diagrams worked. Several of his strains, such as the CIB, are still widely used in *Drosophila* genetics laboratories.

One of Jo's special concerns was the widespread abuse of diagnostic x-ray in medicine and dentistry with the associated hazard to both patients and operators. He gave a principal address at the 3rd International Congress of Human Genetics in Chicago in September, 1966, and his strong interest in the betterment of mankind through the application of genetics continued until his death in the spring of 1967.

The *Drosophila* laboratory at I. U. soon became a major national scientific resource not only for Muller and his students, but for others throughout the world. At first Muller operated it himself with the aid of graduate students and a few technicians. However, the task became so large and complex that a full time position of Research Executive was added. This position was first occupied by *Irwin Herskovitz*, followed successively by *Irwin Oster*, *S. Zimmering*, *W. Lee*, and *R. Reinhardt*, all of whom remain active in teaching and research at various universities and laboratories. *Helen U. Myers* and *Louise Petoe* were long associated with the laboratory. The majority of the *Drosophila* strains developed by Muller and his students are maintained at Bowling Green University, Ohio, at Pasadena, California, and in other laboratories.

Maize Genetics

Since Indiana is definitely in the corn belt of the United States, it is not surprising that a considerable amount of research on maize has been done in the state. I. U., however, was late getting into this field.

Mention has been made already of Paul Weatherwax' pioneer studies on the structure of the flowers in maize. He also encouraged the conservation of the Amerindian stocks of this plant which he considered to be of great value for future studies. He collected these stocks widely in Mexico and South America.

In 1958, *Marcus M. Rhoades* came to I. U. as chairman of the then Botany Department. He had already distinguished himself in cytogenetics of maize in the U. S. Department of Agriculture, at Columbia University and the University of Illinois. He is a member of the National Academy. He brought with him an active research program, and with the able assistance of *Ellen Dempsey* continues his studies in retirement. He and Dempsey have worked extensively on the genetic effects of heterochromatin.

Rhoades' early work at Cornell was on cytoplasmic sterility in maize. He also first recognized preferential segregation and neocentromere formation, and discovered that B chromosomes could induce loss of knobbed members of the regular chromosome complement.

In 1961, Rhoades was instrumental in bringing *Drew Schwartz* to I. U. Schwartz was one of Rhoades' first graduate student at Columbia. Schwartz introduced a new area of maize genetics in the study of isozymes. One of his goals is to determine what regulates gene expression, and how the controlling elements contribute to an organism's development. He continues a program of refined biochemical research on the genetics and other aspects of maize biology. (See also Meserve, 1981, pp. 11-14.)

Developmental Biology, Molecular Biology, and Microbiology

Although pioneer work on the embryology of the armadillo and the badger was done at I. U. by *G. W. D. Hamlett*, some of Eigenmann's work on the eyes of cave vertebrates can be considered developmental, and embryologists *Charles Zeleny*, later at Illinois, was in the Zoology Department from 1904 to 1909, intensive research did not begin until the appointment of *Theodore W. Torrey* to the Zoology faculty in 1932.

Torrey's research was mainly in the field of descriptive and experimental embryology of vertebrates. He studied the degeneration of nerves and sense organs, embryonic sense organs, and especially the embryology and physiology of vertebrate excretory organs. He worked extensively on the early embryogeny of the human kidney and was among the first to recognize that bone preceded cartilage in the evolution of the vertebrate skeleton.

His principal contributions to biology at I. U., however, rest upon his ability as a great teacher and a superb administrator. He was among the first to combine the then conventional comparative anatomy and embryology courses into an integrated developmental course. His textbook on developmental biology was highly successful and is still in print. He was instrumental in the planning of the general biology course for non-majors, and in establishing the Master of Arts for Teachers degree in cooperation with the then Botany Department.

As a departmental administrator *Ted Torrey* ("Terrible Ted" or "T-Square" to the students) had no superior and few peers. His management of Zoology's menage of primadonnas was masterful. He should have written a book on the care and feeding of intellectual tigers. Yet throughout there was never any feeling of neglect among those of us who did not attain international reputations. He was Chairman of Zoology from 1948 until the fall of 1966 when he was succeeded by *W. R. Breneman.*

Following World War II, the phenomenal growth of the university brought need of additional teaching faculty. *Sears Crowell* joined Zoology in 1948, and continues in retirement his research on the morphogenesis of coelenterates here at I. U. and at the Woods Hole Biological Laboratory in the summers. His discovery of stolon fusion in sessile coelenterates and the occurrence of compatible and incompatible strains is noteworthy. He also has used time-lapse photography to study the growth and tissue movements in hydroids.

By 1950, the outlines of the new biology were beginning to emerge, and this was especially apparent in embryology and genetics. Torrey (1968) points out that the physiological and biochemical factors that lie behind visible differentiation and morphogenesis began to be a focus of developmental biology. Attempts were being made to translate such concepts as induction, determination, potency, and gradients into measurable entities. There were hints that nucleic acids were involved in the synthesis of protein, and the first attempts to relate nucleic acids to gene reproduction were being made. Developmental, genetic, and molecular biology were converging. In recognition of the changing direction of research, a search was made for a young biologist in this field.

James E. Ebert joined the Zoology faculty in 1951 and remained until 1956. He did some of his pioneer work on the origin and early function of the vertebrate heart-forming proteins here, and produced a number of outstanding students still active in the field of experimental biology. He left in 1956 to become Director of the Department of Embryology of the Carnegie Foundation in Baltimore where he developed a magnificent program in developmental biology. He is currently president of the Foundation.

Robert W. Briggs came in 1956 upon Ebert's departure. He served as chairman of the Zoology Department from 1969-75. He is noted for his work in collaboration with *T. J. King* on the analysis of changes in embryonic nuclei by nuclear transplantation. At I. U., he and his students have worked on many aspects of embryology using the Mexican axolotl as a primary research organism. He continued his research until his death in 1983.

R(ufus) R(ichard) Humphrey (1892-1977) retired in 1957 after a distinguished career as Professor of Anatomy at the University of Buffalo School of Medicine. He moved his colony of Mexican axolotls and his research program to I. U. and continued working here until his death in 1977. During his long career he studied many aspects of amphibian embryology including anatomy of the testis, interstitial cells of the testis, origin of germ cells, sex reversal, genetics of sex determination, spontaneous and induced polyploidy, and the offspring of polyploids. The axolotl colony with its many strains and mutants has proven to be a valuable aid to research. About 1965 Humphrey produced an axolotl-type albino salamander by hybridizing an albino *Ambystoma tigrinum* with the non-albino white axolotl, *Ambystoma mexicanum.*

The boundary between developmental and molecular biology has become obscure in recent years, and just as those between zoology, botany, and microbiology have already faded, may soon disappear. Most of the workers in the preceding section could also be considered to be molecular biologists. This field has only become self-conscious in the last few years, but parts of it have been around for a long time. In modern biology the boundaries between various areas are definable mainly by the techniques employed so that departmental associations are now largely meaningless.

Salvador E. Luria taught and did research at I. U. from 1943 to 1950. His pioneer work on the genetics of bacteria and bacterial viruses won him the Nobel Prize in 1969.

Irwin C. Gunsalus was briefly at I. U. from 1947 to 1949. He continued his research on microbial biochemistry at the University of Illinois.

Roger Yate Stanier was briefly at I. U. from 1946 to 1947. His research here and at the University of California-Berkeley on the biology of the myxobacteria, bacterial metabolism, and metabolic pathways has won him many honors.

W. Dean Fraser was the last chairman of Microbiology before the departmental fusion. He joined the staff at I. U. in 1955. His research on the morphology, molecular biology, and genetics of bacterial viruses and on the biology of mycoplasma has been outstanding.

Leland S. McClung as mentioned earlier was appointed to the I. U. faculty in 1940 and organized the program in microbiology. He became chairman of the newly established Department of Bacteriology in 1946 and served in that capacity until 1966. He was assistant director of the Division of Biological Sciences from 1965 until 1968. His research has covered many aspects of bacteriology including the taxonomy and ecology of clostridia, food poisoning organisms, bacteriophagy of *Clostridium*, and other aspects of bacterial biology. He continues research at I. U. as Professor of Microbiology Emeritus.

Howard Gest was appointed chairman of the Department of Microbiology in 1966. After retiring from administration he has continued his research on the comparative biochemistry of photosynthetic processes. His work has won him wide recognition. He and *Gary Sojka* form the Photosynthetic Bacteria Group at I. U. (See also Eoyang, 1978, pp. 2-6).

The I. U. molecular biologists persuaded the University early of the need for a specially equipped (P-3) laboratory for research on the transfer of genetic material from one organism to another. This area has created considerable discussion throughout the world, but many of the earlier fears concerning the dangers of such research have abated if not disappeared. Recombinant DNA research may be the key to the solution of many biological problems in several areas.

Endocrinology

In the fall of 1933, *Robert L. Kroc* joined the I. U. Zoology staff to take charge of the beginning biology course and to teach in his speciality, endocrinology. The program which he developed was taken over by *W. R. Breneman* when Kroc left in 1944 to join the research division of Maltine Company of New Jersey (now Warner Lambert).

William R. Breneman received his Ph.D. from I. U. in 1934 and after a National Research Fellowship at Wisconsin and a year of teaching at Miami University returned to I. U. in 1936. His main research interests centered in the endocrinology of the white leghorn chicken, and he has made significant contributions in this field particularly in regard to the pituitary gonadotrophins.

Breneman is noted for his ability as a master teacher and has received several awards. His pre-Christmas lecture "Kalamazoo to You" is deposited in audio in the University archives. The beginning course which he took over from Kroc became a popular and increasingly important core of the undergraduate program in Zoology. In 1966 he became chairman of the Zoology Department and served ably in that capacity until 1969. He retired as Luther Dana Waterman Professor of Zoology in 1977. (See also Meserve, 1981, pp. 4-6.)

General and Systematic Entomology

Among the biologists assembled about *Carl Eigenmann* and *Fernandus Payne* was *Alfred Charles Kinsey* (1894-1956). He occupies a unique position in having achieved prominence in two major fields of biology—entomology and human behavior. He came to I. U. in 1920 after working under *William Morton Wheeler* and *Charles T. Brues* at Harvard.

Kinsey's entomological work represented a distinct departure from that of his predecessors, and he did much to introduce the modern techniques of research in taxonomy in general. His insistence upon massive series, wide geographical coverage, and detailed analyses of every biological aspect of an organism was unique at the time, but has become standard practice in present day taxonomic studies. He was also a pioneer in the introduction of statistics as a tool in systematic studies.

Shortly before World War II, Kinsey became interested in the study of human sexual behavior. The "crusts" which he broke reverberated around the world. Until his death, he maintained that he was a taxonomist applying taxonomic methods to an important social phenomenon. His work continues at I. U. under the direction of *Paul Gebhard*, Director of the Institute of Sex Research which continued to make important contributions in this field. (See also Meserve, 1981, pp. 2-6). Torrey (1965) gives a detailed account of the development of the Institute.

General Protozoology and Protozoan Genetics

Tracy Morton Sonneborn (1905-80) joined the I. U. faculty in 1939, bringing with him his enthusiasm, charisma, and elegant research program. He previously had worked at Johns Hopkins University where he did his graduate studies under *H. S. Jennings*. Sonneborn continued his research at I. U. until shortly before his death. His work centered around the protozoan, *Paramecium aurelia*, in which he studied cellular differentiation, the role of nuclear and cytoplasmic factors in inheritance, serotypes, and mating types. The killer trait was a subject of study for many years, and although it eventually proved to involve a bacterium, its study inspired continuing study of the origin of cellular organelles and other factors.

Tracy Sonneborn was a magnificent lecturer and teacher. One of his greatest contribution to biology was the many students he trained and sent out to teach and do research. Most of them are still active in the field of protozoology.

In 1961, *Mary L. Austin* retired after a long and distinguished career as a teacher and researcher at Wellesley College. She joined the Sonneborn laboratory as a Research Scholar and continues her work on antigenic transformations and other mutations in *Paramecium*.

John R. Preer, Jr. came to I. U. shortly before World War II to work with *A. C. Kinsey* on insects. Kinsey at that time had shifted his interests, and John shifted his interest to protozoan genetics with *Tracy Sonneborn*. After receiving his doctorate in 1947, he moved to the University of Pennsylvania where he distinguished himself in teaching, administration, and research. He returned to I. U. in 1968, bringing as a research associate his wife, *Lousie B. (Bertie) Brandau Preer*, who had received the doctorate from I. U. working with *Ralph L. Cleland* on the cyto-genetics of *Oenothera*. They have continued work on the genetics of protozoa and cell biology of *Paramecium*.

Ruth V. Dippell received her Ph.D. from I. U. in 1950, and joined the Sonneborn laboratory as a research associate. She was appointed to the faculty in 1967, and has continued research and teaching on many aspects of protozoan biology. Her influence extends beyond her teaching and research. Her willingness to cooperate with others and her expertise are always available to students and instructors alike. Her willingness to share cultures has enlivened and enriched many classes.

Myrtle V. Sneller received the M. A. degree from I. U. in limnology in 1949. She has since served as research associate and has done research on mating types, rapid lysis killers, and other aspects of *Paramecium* biology. She was responsible for the maintenance of over 400 genetic strains of *Paramecium* maintained in the laboratory. These strains are now maintained by the National Type Culture Collection.

W. J. van Wagtendonk joined the staff in 1947. He carried on sophisticated experiments on the biochemistry of *Paramecium* until 1965 when he departed for the University of Miami where he recently retired. He developed interesting devices for "purifying" *Paramecium* for chemical studies using the protozoan reaction to an electric current. His major contribution was the development of a defined medium for culturing *Paramecium*.

Ornithology

A general course in ornithology was taught at I. U. for many years, but it was not until *Val Nolan* accepted a joint appointment in biology and the Law School that a research oriented program was introduced. Nolan began studies on the prarie warbler shortly after coming to I. U. in 1949. His monograph published

in 1978 is a masterful analysis of many aspects of the biology of this bird. His recent studies have centered on reproductive strategies and behavior, and broadly on the general ecology and behavior of birds.

Terrestrial Ecology, Evolution, and Ethology

Although aquatic biology developed early at I. U. the teaching and research in general and terrestrial ecology lagged behind. *Stanley Cain* was briefly associated with the botany department in the 1930s but no plant ecology program was ever developed in that field. This seems strange when one considers the seminal influence of the Chicago botanists and their pioneer work on plant succession in the Indiana dunes.

The first formal offerings of ecology were in 1946 and 1947 by *Lamont C. Cole*. As already noted Cole came to I. U. to fill the vacancy in the teaching program caused by the withdrawal of *A. C. Kinsey* into research. Cole developed a general course in ecology the catalog description of which remained unchanged for over twenty years. The contents of the course, however, changed radically during that period.

Charles J. Krebs joined the zoology staff in 1964. He had done his doctoral work in British Columbia and postdoctoral work with *Charles Elton* in England and at the University of California. Krebs developed an active graduate program in the ecology of microtine rodents. His work on population cycles in *Microtus* are now classic, and his successful textbook has done much to encourage the study of the population ecology.

Krebs decided to return to British Columbia in 1966, and *Donald R. Whitehead* joined the botany staff in 1967.

The Division of Biological Sciences

> "It was becoming increasingly evident as early as 1960 that the departmental structure of the biological sciences at Indiana was not appropriately geared to the biology of the times. The prominent research issues of the day were more often biological than zoological or botanical or bacteriological; molecular and genetic biology nurtured by sophisticated biochemistry and the elucidation of the architecture and replicative mechanisms of DNA, were revolutionizing studies of the genetic control of differentiation. A new breed of biologists, few of whom considered themselves botanist, zoologists, or bacteriologists *per se*, was emerging. A new generation of students, conditioned by modernized textbooks, was coming out of high schools. Our conventional departments were being challenged by these circumstances and the simple kind of informal cooperation that had served us well no longer seemed able to meet the challenge. How might we design new types of courses and curricula? How could we recruit faculty personnel who did not identify themselves with conventional disciplines? How could we move frontally toward the acquisition of money, equipment, and laboratory space essential to the advancing frontiers of biology?" (Torrey, 1968).

After much soul-searching, discussion, debate, compromise, and a little acrimony, a charter for a new Division of Biological Sciences emerged. This united the departments of Anatomy and Physiology, Botany, Microbiology, and Zoology. *Tracey Sonneborn* served in 1963-64 as chairman of the executive committee of the Division, and *Charles W. Hagen, Jr.* as Acting Director in 1964-65. *Frank W. Putnam*, a distinguished immunochemist, came in the fall of 1965 from the Univer-

sity of Florida Medical School to be Director with *Leland McClung* as Assistant Director.

Putnam reestablished his laboratory at I. U. and in addition to his administrative duties carried on an active program of research. He has continued teaching and research since his retirements from administration. His work has been basic to the modern understanding of immune reactions. His lab first established the amino acid sequence in the Bence-Jones protein, an immunoglobulin produced by patients with plasma cell tumors. From these studies it is possible to elucidate the principles of antibody structure and specificity. (See also Eoyang, 1980, pp. 11-14.)

Under the divisional organization, the various departments created a variety of courses and programs designed to integrate biology in a modern sense. An instrument center was established to manage expensive equipment such as electron microscopes which could be used by members of any department. Morrison Hall was redecorated to house the expanding faculty and increased numbers of students. Jordan Hall was also extensively remodeled to provide for new types of equipment and laboratories. Approval was finally obtained in 1981 to go ahead on the long planned addition to Jordan Hall.

In 1969, another distinguished biochemist, *C. H. Werner Hirs* became director of the Division. He too carried on an active program of research on the structure, function, and chemistry of proteins and glycoproteins. In 1977 Hirs moved with his laboratory to the University of Colorado Medical School.

During the tenure of Putnam and Hirs the faculty in all departments increased, and many new and innovative research programs were established. The pattern of courses and the laboratory equipment were also modernized so that a student today can work in a wide variety of areas or even in the interstices between areas.

The Present Biology Department

In 1977 it was decided to abandon the divisional organization and integrate the departments (with the exception of Anatomy and Physiology) in a single Department of Biology. *John Preer* became the chairman from 1977 to 1979, and *Gary Sojka* was chairman from 1979 to 1981. *Albert L. Ruesink* served as acting chariman (1981-82). *Rollin C. Richmond* is currently chairman.

Epilogue

Historians more often are rewarded by criticism of their omissions than their commissions. I have tried to make this brief history as complete and informative as possible to reveal the pathways down to the present and the present breadth and direction of research. I have largely neglected teaching which has been an intrinsic part of the whole process, but almost without exception teaching at the upper undergraduate and graduate levels has closely followed research.

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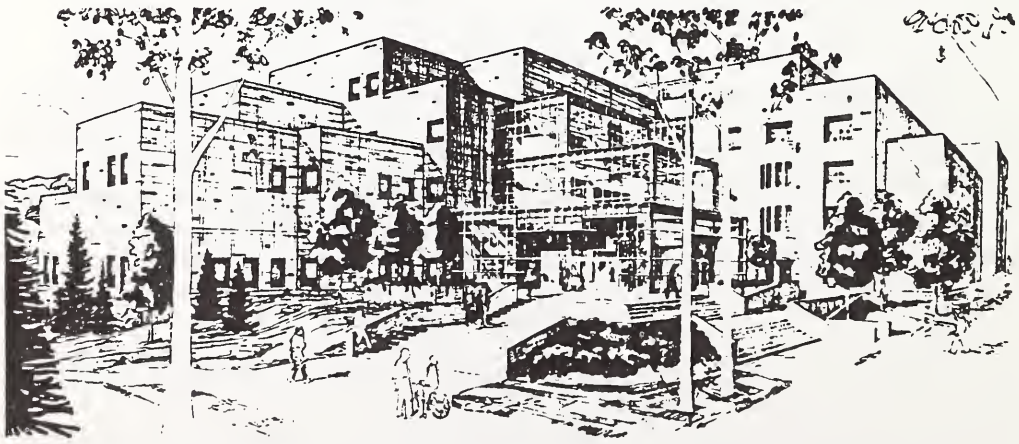

New Addition to Jordan Hall's Department of Biology — Now under construction.

MICROBIOLOGY AND MOLECULAR BIOLOGY

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Indiana University School of Medicine
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Department of Biology
Indiana University-Purdue University at Fort Wayne
Fort Wayne, Indiana 46805

ABSTRACTS

Possible Aerobic Nitrate Reductase Activity in Paracoccus denitrificans. WILLIAM W. BALDWIN, W. MARSHALL ANDERSON and MING TSENG, Northwest Center for Medical Education, Indiana University School of Medicine, Gary, Indiana 46408.——*P. denitrificans* has a doubling time of 200 min. when grown aerobically on a minimal salts-nitrate-glucose medium or anaerobically on the same medium. When grown aerobically on a nutrient broth-glucose (NBG) medium the doubling time was 160 min., but decreased to 80 min. with added nitrate. Similar results were obtained if succinate was substituted for glucose. The anaerobic doubling time was 120 min. on the NBG-nitrate medium. It is generally assumed that in *P. denitrificans* O_2 inhibits nitrate reductase activity and represses its synthesis. To determine the role of nitrate reductase in aerobic growth, mutants lacking the activity (approximately 1/50 parental activity) were isolated by primary selection for chlorate resistance and tested for ability to grow anaerobically on nitrite but not grow on nitrate containing medium. One mutant selected for further study had an aerobic doubling time in NBG nitrate medium identical to the parental strain grown aerobically without nitrate. Revertants of the mutant are being isolated to define further the role of nitrate reductase in aerobic growth. It is postulated that nutrient broth may contain either a substance preferentially oxidized by nitrate reductase or a cofactor that protects the enzyme from inhibition and/or repression by O_2. (Supported by Lake County (IN) Medical Center Development Agency).

Detection of ϵ-N-Methyllysines, in Mucor racemosus, by High-Performance Liquid Chromatography. J. R. GARCIA, Ball State University, Muncie, Indiana 47306 and P. S. SYPHERD, University of California at Irvine, Irvine, California.——This study was undertaken in order to test the possibility of using high-pressure liquid chromatography (HPLC) for the detection and resolution of methylated lysine residues. While HPLC has been used extensively for the detection and resolution of unmodified amino acids, it hasn't been employed in work with methylated amino acids. Dansylated derivatives of ϵ-N-mono, ϵ-N-di, and ϵ-N-trimethyllysine (MML, DML, and TML, respectively) standards were well resolved from each other and the rest of the amino acids, commonly found in proteins, through the use of a reversed-phase LDC Spherisorb column. The separation was achieved using a 0-60% concave methanol gradient and the fluorescence of the effluent monitored through the use of a fluorometer. In order to test the technique with actual biological samples, proteins known to contain methylated amino acids were acid hydrolyzed, dansylated and examined. MML, DML and TML, which had previously been tentatively demonstrated in the alpha subunit of *Mucor racemosus*

protein synthesis elongation factor-1, were all detected and well resolved. In addition, the TML in the cytochrome c of yeast was also detected. In conclusion, a rapid and rather sensitive HPLC technique has been developed which allows for the detection as well as resolution of MML, DML, and TML in hydrolyzed protein samples.

Suppression of Bacterial Endotoxin (LPS) Mitogenicity for Spleen Cells by Tumor Cell Culture Fluids. H.M. GHARPURE and K.W. BRUNSON, Northwest Center for Medical Education, Indiana University School of Medicine, Gary, Indiana 46408.——Sequential monitoring of mitogen induced proliferative responses of spleen cells from animals bearing a less metastatic RAW117-P lymphosarcoma or its highly metastatic variant RAW117-H10 lymphosarcoma tumor revealed that responses to either the B cell mitogen, lipopolysaccharide, or the T cell mitogen, concanavalin A are not affected until tumor-bearing day 13. Spleen cells from mice bearing the more metastatic tumor RAW117-H10 selectively suppressed the Con-A but not LPS induced proliferation of normal spleen cells. Direct co-culture of either of these two tumor cell lines with normal spleen cells showed that proliferation of normal spleen cells to the T cell mitogen Con-A is enhanced by the parental (less metastatic) tumor. In contrast the more metastatic tumor RAW117-H10 suppressed the Con-A response. Responses to LPS were inhibited by both RAW117-P and RAW117-H10 tumor cells. This mode of suppression was not restricted to spleen cells from syngeneic animals, but also affected spleen cell response from outbred mice. Serum free culture supernatants from both the tumor cell lines inhibited LPS induced proliferation of B cells from normal spleen. However, the supernatant enhanced Con-A induced proliferation. Increasing experimental evidence suggests a dualistic role of the culture supernatants in achieving differential effects on distinct spleen cell populations. (Supported in part by the funds from the Lake County Medical Center Development Agency).

Transfer of the Plasmid RPI into the Chemolithotrophic Bacterium, Thiobacillus Neapolitanus. CHARLES F. KULPA, MICHAEL T. TRAVIS, MARK T. ROSKEY, Department of Microbiology, University of Notre Dame, Notre Dame, Indiana 46556.——RPI, an incompatibility group PI plasmid specifying multiple drug resistances, has been transferred into the chemolithotroph *Thiobacillus neapolitanus*. The ability of *T. neapolitanus* to receive, express, and transmit RPI encoded antibiotic resistance is examined. We have repeatedly transferred RPI by conjugation from *Escherichia coli* ROE53I to *T. neapolitanus*, with a frequency of approximately 8.5 X IO^{-5}. The transfer of RPI into *Pseudomonus aeruginosa* from *E. coli* ROE531 yielded similar frequencies (4.8 X IO^{-5}). Reciprocal matings with strains of *T. neapolitanus* (RPI) derived from a prior cross, yielded transfer frequencies into ROE53I and PAO2 of 7.9 X IO^{-7} and 6.0 X IO^{-7}, respectively. Additionally, the transfer frequency has been shown to be dependent on the donor to recipient ratio; higher transfer frequencies occurring at decreased donor to recipient ratios. The presence of the RPI plasmid in the transconjugants was shown by isolation of RPI plasmid DNA and subsequent characterization by agarose gel electrophoresis. The presence of RPI DNA was detectable in both the *T. neapolitanus* (RPI) transconjugants and those strains derived by reciprocal mating. These data indicate that the RPI plasmid host range extends beyond heterotrophic bacteria, and RPI can be expressed in a lithotrophic bacterium totally unrelated to its previous host.

Slime Producing Bacteria Involved in Urinary Tract Infections. LYNN A. LARUE, WAYNE J. LENNINGTON and DONALD A. HENDRICKSON, Ball State University, Mun-

cie, Indiana 47306.——Urinary tract infections are one of the most commonly encountered bacterial infection found in hospital laboratories. The species, strains, and number of organisms identified at Ball Memorial Hospital, Muncie, Indiana, as the causative agent of a urinary tract infection were classified by API 20 strips or microscan plates. Special attention was shown to organisms producing a glycocalyx. Negative capsule stains were used to distinguish between capsule producing and non-capsule producing bacteria. Ten different species of organisms were identified, with *Escherischia coli* being the most abundant and most varied, having thirty-three different strains. All organisms were capsular, except for three strains of *E. coli* and one strain of *Pseudomonas aeruginosa*. The role of the glycocalyx in this infection could not be determined, but further research is being conducted to investigate this relationship. Research is continuing in order to find out if the capsule is a characteristic of this infection or if it has appeared in our data due to a new factor in this area selecting for capsular bacteria.

Fungal Culture Technique. CAROL A. WEAN and DONALD A. HENDRICKSON, Ball State University, Muncie, Indiana 47306.——A technique for the culture and microscopic examination of fungi has been devised in order to overcome inadequacies present in other methods which require a transfer from the culture plate to the microscopic slide. In this technique the fungus is cultured directly on the microscope slide, thus eliminating the transfer process.

To avoid contamination during incubation, a culture chamber is established by placing a layer of absorbant paper in the bottom of a glass Petri plate. A bent glass rod is centered in the plate, upon which a glass concavity (hanging drop) microscope slide is autoclaved.

After the chamber has cooled, sterile water is added to absorbant paper. Sterile Sabouraud dextrose broth is lightly inoculated with a fungal culture using an inoculating needle. The cover is replaced and the culture allowed to incubate undisturbed. When the culture has reached the appropriate growth stage, a cover slip is positioned over the concavity and the slide examined microscopically.

This culture technique is simple and uses materials commonly found in microbiology laboratories. The resulting slides should be beneficial for teaching purposes.

Effect of Dietry Tannic Acid on the Facultative Gut Microflora of the Red Squirrel

KARA EBERLY and ERIC MOULD
Department of Biology, Saint Mary's College, Notre Dame, Indiana 46556
and
DOUGLAS DUFF
Department of Biology
Indiana University, South Bend, Indiana 46334

Introduction

The polyphenolic compounds (tannins) have been the subject of considerable study because of their action in inhibiting ruminant digestion when they are present in feeds(1,11). Polyphenolics have also been reported to inhibit digestion in monogastric animals. However, the native diets of some animals, including the red squirrel, are high in polyphenolic compounds. In digestion studies with red squirrels fed laboratory diet supplemented with tannic acid, one of the less toxic polyphenolics, dry matter and lipid digestion were found to be unaffected by up to 5 percent dietary tannic acid after a two week period of adaptation to the diet(12). This ability to adapt to a high tannic acid diet could be the result of changes in enzymatic levels in the squirrels, changes in the gut microflora, or both.

The well-documented sensitivity of many bacteria to tannic acid, as well as several reports of bacteria and fungi which can tolerate tannic acid or even use it as a carbon source(3-11,13-16,18,19), led to the examination of the effects of dietary tannic acid on gut microflora. If the gut microflora were essentially unchanged in quantity and composition, then a bacterial role in adaptation to dietary tannic acid would be highly unlikely. Changes in the microflora might reflect sensitivity to tannic acid without any effect on digestion. In this case, however, microflora composition could prove to be indicative of the animal's natural diet at the time of evaluation. On the other hand, floral changes could be highly significant to the squirrel's ability to utilize tannic acid containing diets.

Fecal samples were chosen because bacteria from the whole gastrointestinal tract were of interest; fecal samples should contain bacteria from the stomach and small intestine as well as colon organisms; and a series of diets could be tried on the same animals. Total bacterial counts were chosen to evaluate the overall effects on gut bacteria since they should reflect primarily populations of anaerobes. Finally, a series of selective and differential media were chosen to quantitate the populations of the predominant facultative organisms.

Materials and Methods

Red squirrels (*Tamiasciurus hudsonicus*) were live trapped and housed in 45 cm by 45 cm by 25 cm metal cages on the same cage rack. Thus there was no direct contact between animals, and opportunities to coprophagy were reduced. However, there was no attempt to strict isolation, nor was the diet presterilized. Squirrels were fed *ad libitum* on a balanced rodent ration from Bio-Serv Inc. which contained 18.5% protein, 5.05% fat, 2.99% ash. The diet was supplemented with 0, 0.5, 0.75, 1.0, 2.0, or 5.0 percent tannic acid which replaced undigestible fiber. Tap water was provided *ad libitum*.

Fecal samples were collected within 24 hours after capture and after 7 days on the given diet. Other collection times are indicated in the results. Samples

were aseptically weighed and uniformly suspended in sterile saline at a 1/100 dilution. Further ten-fold dilutions were made in trypticase soy broth and plated on trypticase soy agar (TSA), KF streptococcal agar(KF), Mitis-salivarius agar(MI), Levine's eosin methylene blue agar(EMB), Salmonella-shigella agar (SS), Mannitol-salt agar (MS), and Rogosa SL agar(SL). After plates were incubated 48 hours at 37C in candle jars, characteristic colonies were selected, gram-stained, restreaked, and identified as to group by conventional bacteriological methods. A sample of bacteria diluted 1/100 in saline was preserved in ten percent formalin and counted on a Petroff-Hauser counter for total counts. A total of 9 squirrels were evaluated in three different experiments. In two experiments increasing amounts of tannic acid were fed sequentially. In the third experiment the control diet was followed immediately by five percent tannic acid. Data were analyzed using a one-way analysis of variance.

Results

Direct bacterial counts on squirrels which had received the control diet for 7 and 13 days averaged 3.5×10^{11} bacteria per gram of feces. Total counts averaged 4.2×10^{11} per gram feces on days 1 and 7 after a change to a 5% tannic acid diet and 3.5×10^{11} per gram feces on days 2 and 7 after a return to the control diet. There were no statistically significant changes in direct counts. Total facultative counts for the control diet at 7 and 13 days averaged 8.5×10^{8} or .04 % of the total count. Therefore, the direct counts represent chiefly anaerobes, and the total anaerobe population was not significantly altered by the tannic acid in the diets.

Quantitative changes were observed in the facultative gut microflora. There was approximately a 1000 fold decrease ($p < .05$) in the counts of the genus *Lactobacillus* when tannic acid was included in the diet (Table 1 and Figure 1).

Gram negative enteric bacilli, staphylococci, yeasts, and enterococci were also isolated. However, there was no statistically significant relationship of their population sizes to tannic acid in the diet (Table 2). The mean log enteric count on the control diet was 6.9 with a standard deviation of 1.24. However, patterns with individual animals were quite variable; 4 squirrels showed moderate to extensive decreases with increased tannic acid levels; 1 showed a progressive increase; and 3 showed no pattern at all with wide fluctuations in enteric levels. Four squirrels failed to have detectable levels (greater than 1000/g feces) of enterococci; 2 only developed them at the 5% tannic acid level; 1 had an erratic pattern; and 2 showed moderate increases in enterococcus levels. Variations among animals were also seen with the staphylococci and yeasts. Although there was

TABLE 1. *Fecal Lactobacillus counts of red squirrels fed artificial diets with variable tannic accid content for a period of 7 days.*

DIET % Tannic Acid	LOG VIABLE COUNT[a] Per Gram Feces (Mean ± standard dev.)	NUMBER
0	7.90 ± 0.54	6
0.5	5.51 ± 1.01	3
0.75	6.10 ± 0.84	6
1.00	5.44 ± 1.71	6
2.00	5.23 ± 0.47	5
5.00	4.70 ± 1.42	8

[a] *Lactobacillus* counts were made from MI, KF, and TSA plates, which gave consistently higher counts than Rogosa SL medium.

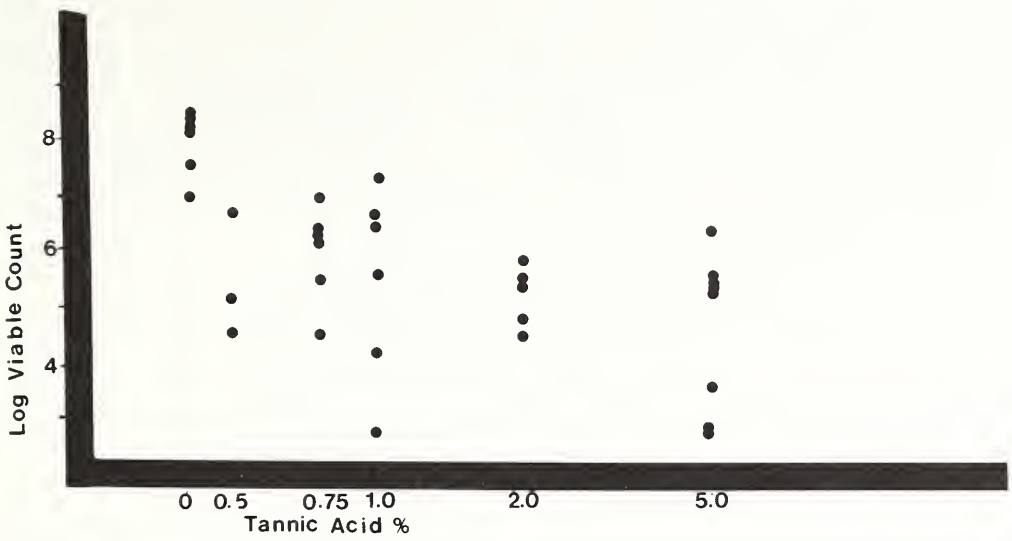

FIGURE 1. *Seven-day fecal Lactobacillus counts of red squirrels fed artificial diets with variable tannic acid content.*

no decrease in population levels of staphylococci or yeasts in squirrels with detectable populations (1000 or more), there appeared to be a tendency towards reduced numbers of animals with detectable levels in the presence of higher dietary tannic acid levels (Table 2). However, these reductions were not statistically significant at the 5% level (Chi square test).

One important question concerned whether the results obtained at 7 days on a diet represented a period when lactobacillus counts were still falling or a time at which they had reached their lowest levels. Secondly, we needed to know how rapidly populations returned to control levels. Lactobacillus counts were made at 7 and 13 days on the control diet; then animals were switched to a 5% tannic acid diet, and samples were assayed on days 1 and 7 on the 5% diet. Finally, animals were returned to the control diet and samples were assayed on day 2 (Table 3). It appears that the 7 day sample underestimates slightly the initial effect of tannic acid on levels of fecal lactobacilli but does provide a reasonable time point for comparing the effects of various dietary tannic acid levels. Recovery of the population levels appeared to be essentially complete in 2 days.

Finally, comparisons between population levels at capture and after 7 days on the control diet showed only 2 instances where an organism which had not been present in detectable levels at capture became detectable on the new diet. A staphylococcus reached population levels of 1.8×10^5 in a squirrel which had had less than 10^3 /g feces at capture, and a squirrel with undetectable lactobacillus levels at capture reached populations of 1.5×10^8 / g. Populations of enterics and lactobacilli each increased in 5 of 6 squirrels when they were put on the control diet.

Discussion

Polyphenolic compounds appear to be important plant defenses against herbivores, and digestibility by herbivores is often reduced by high phenolic content (1,11). Native squirrel diets, however, contain large amounts of phenolic compounds. In controlled feeding studies using tannic acid supplemented diets, reduced dry matter and lipid digestibility were observed on the first diet fed (0.5% tannic acid), but then the animals appeared to adapt to tannic acid, and digestibility on the subsequent diets (0.75% to 5.0% tannic acid) returned to control levels (12).

TABLE 2. *Facultative fecal counts of red squirrels fed artificial diets with variable tannic acid content for a period of 7 days.*

DIET % Tannic Acid	PREVALENCE *Detectable* Total	LOG VIABLE COUNT Squirrels with counts only (/g feces)
GRAM NEGATIVE ENTERIC BACILLI[a]		
0	6/6	6.90 ± 1.24
0.5	3/3	3.95 ± 0.98
0.75	4/6	6.27 ± 0.72
1.0	5/6	6.25 ± 1.29
2.0	2/5	6.21 ± 1.77
5.0	6/8	6.81 ± 1.36
ENTEROCOCCI[b]		
0	3/6	5.98 ± 0.23
0.5	2/3	3.76 ± 1.07
0.75	2/6	6.04 ± 0.00
1.0	2/6	5.51 ± 0.99
2.0	1/5	3.30 ± 0.00
5.0	3/8	4.25 ± 1.23
STAPHYLOCOCCI[c]		
0	6/6	4.49 ± 0.54
0.5	2/3	5.52 ± 0.34
0.75	4/5	4.26 ± 1.07
1.0	3/6	3.85 ± 0.81
2.0	4/5	4.30 ± 0.86
5.0	3/8	5.14 ± 0.43
YEAST[d]		
0	3/6	5.86 ± 0.97
0.5	1/3	4.30 ± 0.00
0.75	3/5	5.66 ± 1.33
1.0	1/6	5.51 ± 0.00
2.0	0.5	— —
5.0	2/8	7.31 ± 0.30

a. Enteric counts were made from EMB and TSA plates.
b. Enterococcus counts were made from KF and MI plates.
c. Staphylococcus counts were made from MS plates.
d. Yeast counts were made from TSA, EMB, MI, and RSL plates.

One of the frequently reported actions of tannic acid is to bind to proteins thus reducing protein digestibility and inactivating enzymes (11). Therefore, the adaptation to dietary tannic acid could involve the increased secretion of digestive enzymes by squirrels. However, a number of aerobic and facultative bacteria and fungi have been reported which can degrade tannic acid, and some can use it

TABLE 3. *Fecal Lactobacillus counts of red squirrels fed artificial diets containing 0% and 5% tannic acid for varying time periods.*

DIET % Tannic Acid	DAY	LOG VIABLE COUNT[a] Per Gram Feces (Mean ± Standard div.)	NUMBER
0	7	8.24 ± 0.07	3
5	1	4.76 ± 1.87	3
5	7	5.78 ± 0.53	3
0	2	7.73 ± 0.22	3

[a] *Lactobacillus* counts were made from MI, KF, and TSA plates, which gave consistently higher counts than Rogosa SL medium.

as a sole carbon source (4-11,14,18,19). Therefore it is reasonable to examine the population levels of yeasts and bacteria in the gastrointestinal tract to determine their potential involvement. Bacteria have often been reported to vary in their sensitivity to tannic acid, so that even if they do not participate in tannic acid degredation, they may serve as indicators of dietary tannic acid (or polyphenolic levels). Fecal samples allow us to monitor bacteria from various parts of the gastrointestinal tract and of course permit repeated sampling from the same squirrel.

Total direct bacterial counts were made to determine if there were a quantitative effect on the overall anaerobe population, and none was found. The anaerobe counts, approximately 10^{11} /g of feces, are similar to those reported for rats, mice, and ground squirrels (2,17). This does not rule out changes in composition of the anaerobic flora; in fact such a change would be expected.

The facultative organisms isolated belonged to groups commonly found in the gastrointestinal tract and feces of mammals. Not all groups were represented in all squirrels at capture, which is not unexpected. The use of separate cages apparently prevented the free exchange of microflora. Therefore, the pattern of change in floral levels with increased dietary tannic acid often varied with the squirrel. Studies are presently underway in this laboratory to see if these differing patterns are due to differences in sensitivity of the isolates to tannic acid and/or their ability to utilize tannic acid as a carbon source.

Only *Lactobacillus* populations showed a significant decrease in the presence of dietary tannic acid. This was anticipated since gram positive organisms have been reported to be more sensitive to tannic acid than gram negatives (18), and that has been confirmed in our laboratory. Also, the lactobacilli are found predominantly in the mammalian stomach(17), and that would be the area of highest exposure to tannic acid. Timed studies were done in an attempt to confirm this finding; and in fact lactobacillus populations were lower in all squirrels on day 1 after introduction of tannic acid into the diet than on day 7. This was not found to be the case with the other facultative microbes. Although we cannot say that day 7 represents a steady state condition, it does appear to represent a reasonable sampling time. Recovery of the *Lactobacillus* populations was essentially complete two days after return to the basal diet. Perhaps this is related to the reduced digestibility of the 0.5% tannic acid diet. If dietary tannins are responsible for a decrease in fecal lactobacilli, one could predict that freshly captured squirrels would have lower counts than after one week on the basal diet, and this was found to be the case.

In general, changes in dietary tannic acid levels did cause perturbations in the facultative gut microflora while overall anaerobe levels remained essentially constant. These changes may or may not be related to the ability of the squirrels to adapt to tannic acid in the diet. However, changes in lactobacillus counts do appear to be related to dietary tannic acid levels, and perhaps to the levels of other tannins.

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Effects of the Antibiotic Tunicamycin on Murine Lymphosarcoma Cells.

S. S. Joshi, K. E. Tsaparikos, J. Loh and K. W. Brunson
Northwest Center for Medical Education
Indiana University School of Medicine
Gary, Indiana 46408

Introduction

Metastasis, the spread of tumor cells from the site of origin to other distant sites, is a complex biological phenomenon and depends on both host and tumor cell properties (1, 5, 10). Several experimental model systems have been developed to study some of the various aspects of cancer metastasis (4, 11). One such model system, a murine lymphosarcoma, was developed by Brunson and Nicolson (2) using the Abelson leukemia virus induced murine lymphosarcoma RAW 117 cell line. A highly malignant and liver metastatic variant line RAW 117-H10 was derived from the parental less malignant line RAW 117-P by using an *in vivo* selection method of cycles of intravenous inoculation of tumor cells and harvesting solid liver tumors in syngeneic BALB/C mice. The metastatic variant RAW 117-H10 line differed from the parental tumor cell line in that the RAW 117-H10 cells formed about 100-200 times more liver tumor nodules than he parental line RAW 117-P, expression of viral glycoprotein component gp70 was decreased on the RAW 117-H10 cells when compared to the parental line RAW 117-P and the highly malignant cells bound less concanavalin A lectin than their parental counterpart RAW 117-P (12).

It was also shown in our laboratory that there are two major antigens expressed on these cell lines as identified by xenoantisera and that expression of antigens on the two cell lines are different (3, 7). One antigen is expressed more on the parental line RAW 117-P and distributed uniformly over the cell surface. A second antigen is expressed more on the highly malignant RAW 117-H10 cells than the less malignant RAW 117-P cell line and the distribution of this second antigen is of a patchy nature (Joshi and Brunson, unpublished). These two antigens have been characterized as glycoproteins which are displayed on lymphosarcoma cell surfaces.

Although distinct changes in the cell surface properties associated with neoplastic transformation is well documented, the exact relationship of cell surface properties and cancer metastasis is poorly understood. In the present study the antibiotic tunicamycin, a known glycosylation inhibitor, was used to study glycosylation inhibition effects on growth, glycoprotein synthesis, and both *in vitro* and *in vivo* biological behavior of these tumor cells.

Materials and Methods

Cell Culture

Both the RAW 117-P and RAW 117-H10 cells were maintained in 60mm x 10mm petri dishes (Falcon 1007, Falcon Plastics, Oxnard, California) containing Dulbecco's Modified Eagle Medium (DMEM) with 10% bovine calf serum (Flow Laboratories, McLean, Virginia) and no antibiotics.

Growth Inhibition Assay

5×10^2 cells in DMEM containing 2% bovine calf serum (BCS) were plated in each well (0.1ml/well) with or without different concentrations of tunicamycin

in microculture tissue culture plates (Linbrow, Flow Laboratories). Tunicamycin (Sigma Chemical Co., St. Louis, Missouri) stock solution was prepared by dissolving 2mg of the drug in 1 ml 10mM NaOH, diluted to the required concentration with DMEM and sterilized by filtering through 0.45 filters (Gelman, Ann Arbor, Michigan). Six hours before harvesting the cells 0.5 μCi of ^{3}H Thymidine (ICN Chemicals, Irvine, California) was added to each well. Cells were harvested at 24 hour intervals using a "Mini Mash" cell harvester (MA Bioproducts, Walkersville, Maryland). The cells harvested in the Mini Mash device on glass fiber filter papers were counted in a liquid scintillation counter (Beckman model LS230) to determine tritiated thymidine incorporation.

Measurement of Glycoprotein Synthesis

Glyoprotein synthesis was measured by culturing 5 x 10^3 cells/ml (5 ml total volume 60mm petri dish) in DMEM containing 2% BCS, and ^{3}H-fucose (New England Nuclear, Boston, Massachusetts) was added 6 hours before harvesting the cells. Procedures for harvesting and counting the radioactivity were essentially as described above for tritiated thymidine incorporation.

Measurement of Adherent Cell Populations

5 x 10^3 cells per ml (total 10 ml) were plated in 100mm x 15 mm (1001 Falcon) petri dishes with or without the appropriate amount of tunicamycin. Cells were then incubated for 36 hours at 37°C in an atmosphere containing 5% Co_2 and 95% air. After the incubation period nonadherent (floating) cells were first carefully removed with a pipette, and any settled nonadherent cells were removed by immediate gentle washing of the dish with two volumes of 5 ml each DMEM. Five ml of 0.02 M EDTA (Sigma Chemical Co.) in calcium-free phosphate buffered saline, pH 7.2, was added to each dish and allowed to remain at 37°C for 5 min. Adherent cells were then harvested by vigorous pipetting. Cell suspension samples were counted and the percentage of adherent cells in each total population was determined.

In Vivo Experiments

Biological properties of the tunicamycin treated (36-hr incubation, 37°C in 5% Co_2) and untreated cells were assayed by inoculating 5 x 10^3 cells in 0.1 ml of serum-free DMEM intravenously (tail vein) into syngeneic BALB/C mice. Death of the animals were noted daily following injection of tumor cells.

Results and Discussion

Figure 1 shows the growth inhibitory effects of tunicamycin at various concentrations on the less malignant parental line RAW 117-P. At least 1 μg/ml concentration of tunicamycin was necessary for significant growth inhibition of these cells compared to control (no tunicamycin) cell cultures. A dose of 1 μg or more tunicamycin per ml completely inhibited the growth of the RAW 117-P cells for as long as 72 hours after addition of the drug. Interestingly, the tunicamycin concentration (1 μg/ml) which inhibited growth of RAW 117-P cells did not exhibit the same effect on highly malignant RAW 117-H10 cells. Even a tunicamycin concentration of as much as 2.5 μg/ml did not have any significant growth inhibitory effect on the RAW 117-H10 cells (Fig 2). As shown in Fig 3, a dose of at least 5 μg/ml tunicamycin concentration was necessary to exert a growth inhibitory effect on the highly metastatic variant cell line RAW 117-H10 similar to that seen with the RAW 117-P cells at 1 μg/ml.

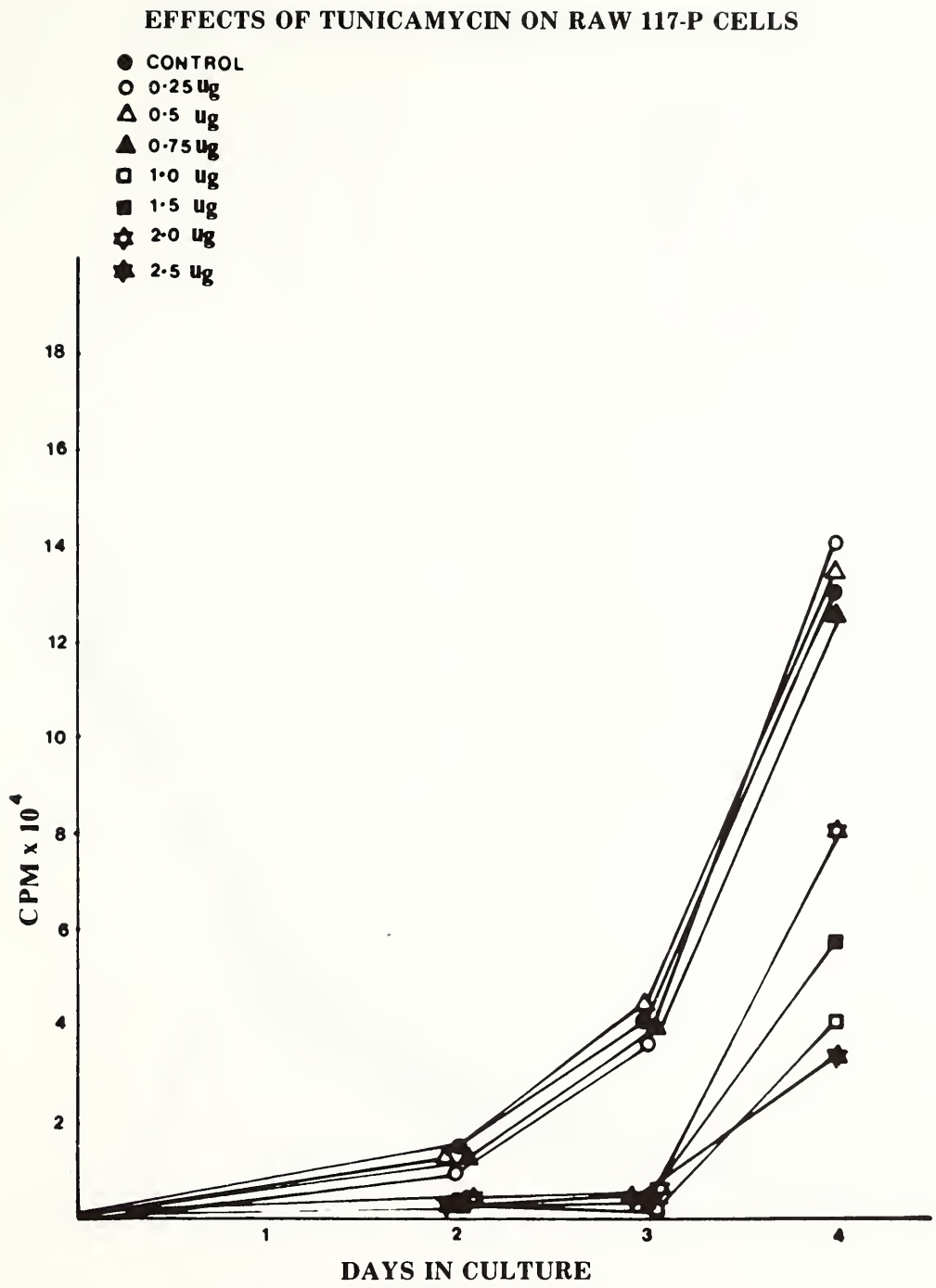

FIGURE 1: *5 x 10² RAW 117-P cells were plated in 0.1 ml volume in each well in microculture plates and incubated at 37°C in 5% Co_2, with or without an appropriate concentration of tunicamycin. Six hrs. before harvesting the cells, 0.5 μ Ci of ^{3}H-thymidine was added. Each point represents the average of triplicate determinations (Cpm).*

As expected, tunicamycin was shown to have an inhibitory effect on glycoprotein synthesis in these cells, as measured by ^{3}H-fucose uptake. As shown in Figure 4, at a concentration of tunicamycin of 1 and 2 μg/ml there is inhibition of glycopro-

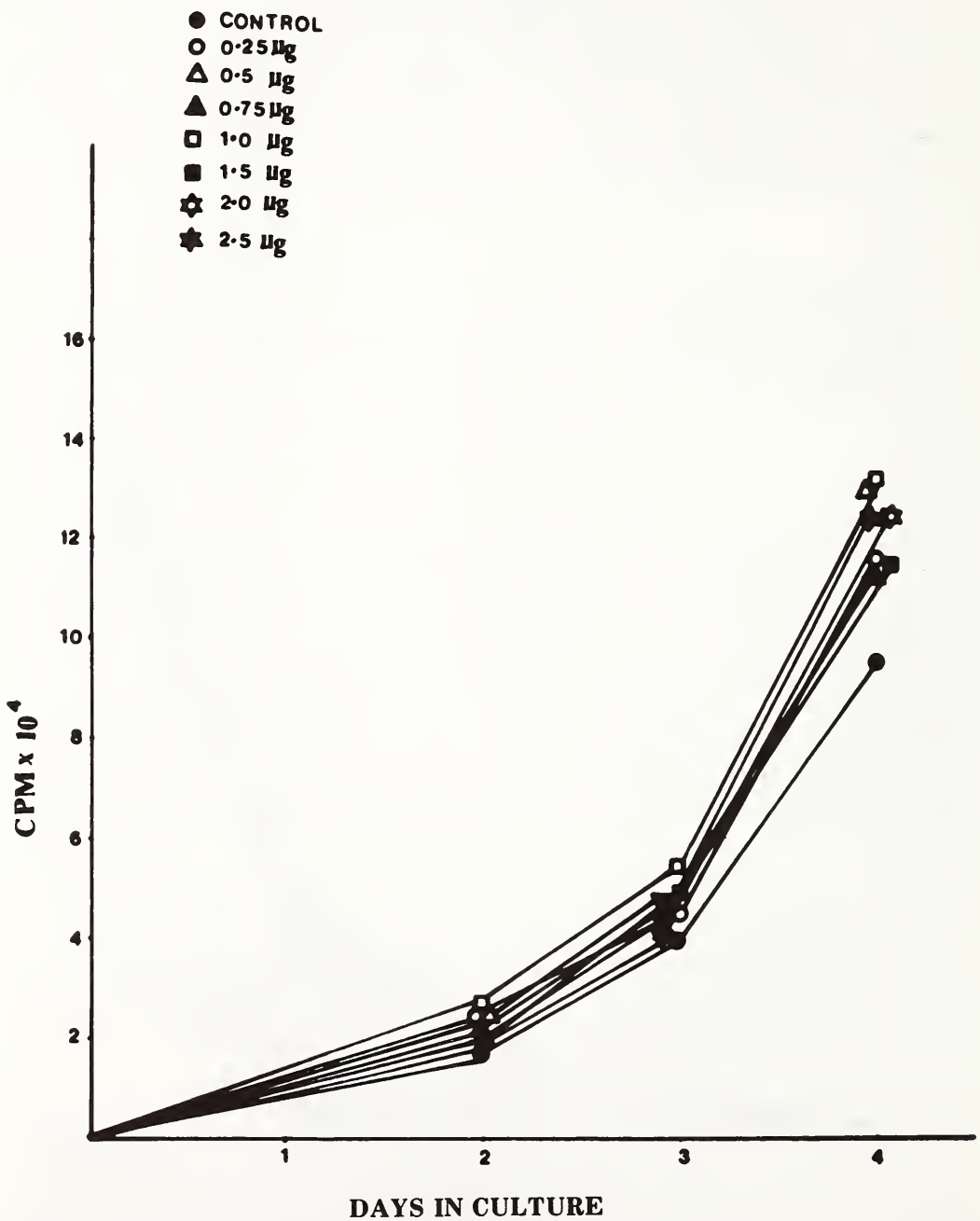

FIGURE 2: *5 x 10² RAW117-H10 cells were plated in 0.1 ml volume in each well in microculture plates and incubated at 37°C in 5% Co_2, with or without an appropriate concentration of tunicamycin. Six hours before harvesting the cells, 0.5 µ Ci of ^3H thymidine was added. Each point represents the average of triplicate determinations (Cpm).*

tein synthesis of about 30% and 50%, respectively, in the less malignant RAW 117-P cells. The highly malignant RAW 117-H10 cells showed only a slight inhibition of glyprotein synthesis with 1 and 2 µg/ml concentration of tunicamycin (approximately

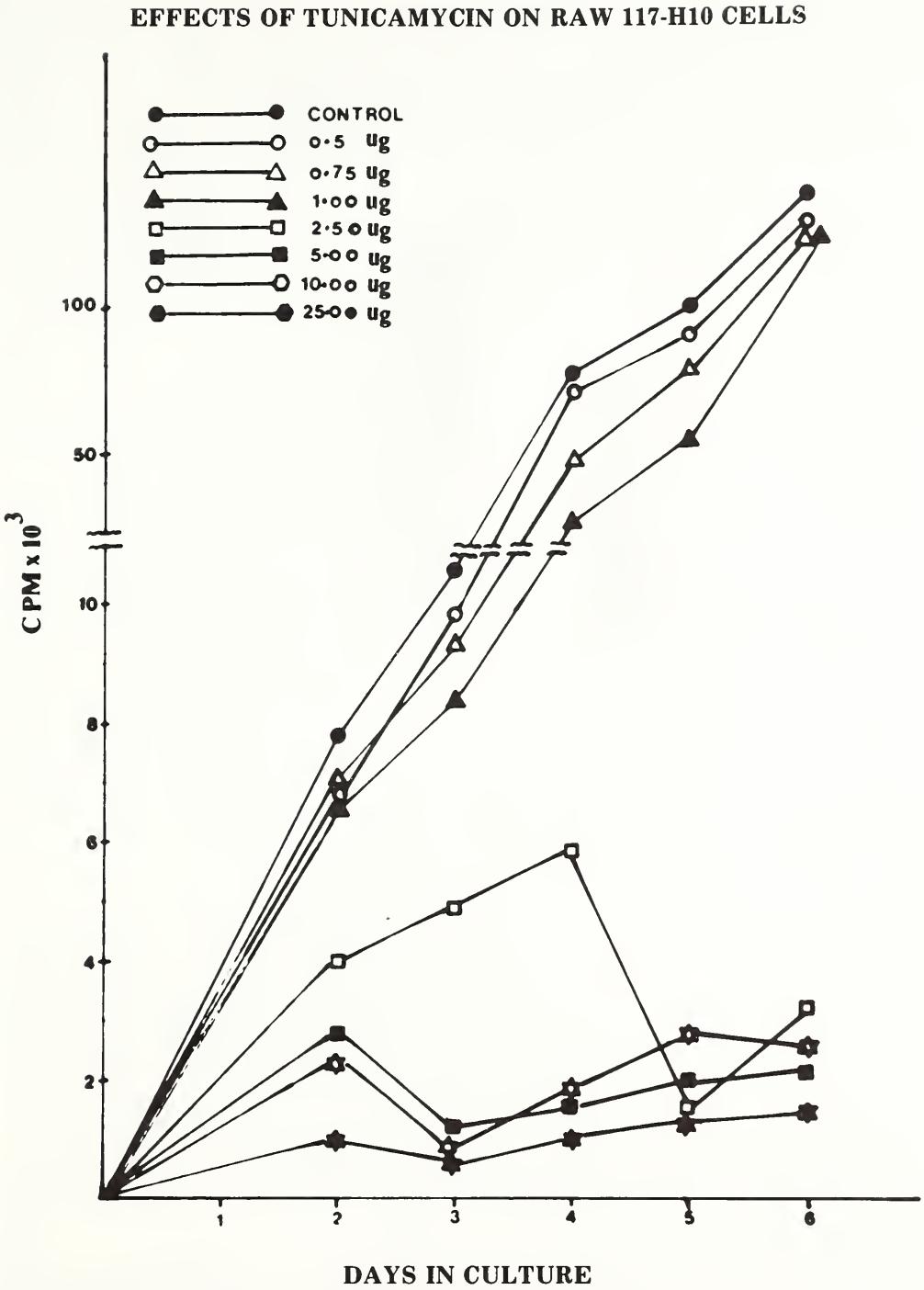

FIGURE 3: *5 x 10² RAW117-H10 cells were plated in 0.1 ml volume in each well in microculture plates and incubated at 37°C in 5% Co_2, with or without an appropriate concentration of tunicamycin. Six hours before harvesting the cells, 0.5 μ Ci of ^3H thymidine was added. Each point represents the average of triplicate determinations (Cpm).*

10% inhibition at 2 μg/ml) although higher concentrations did show similar inhibition of glycoprotein synthesis (data not shown).

Tunicamycin was also shown to affect the adherent properties of these lym-

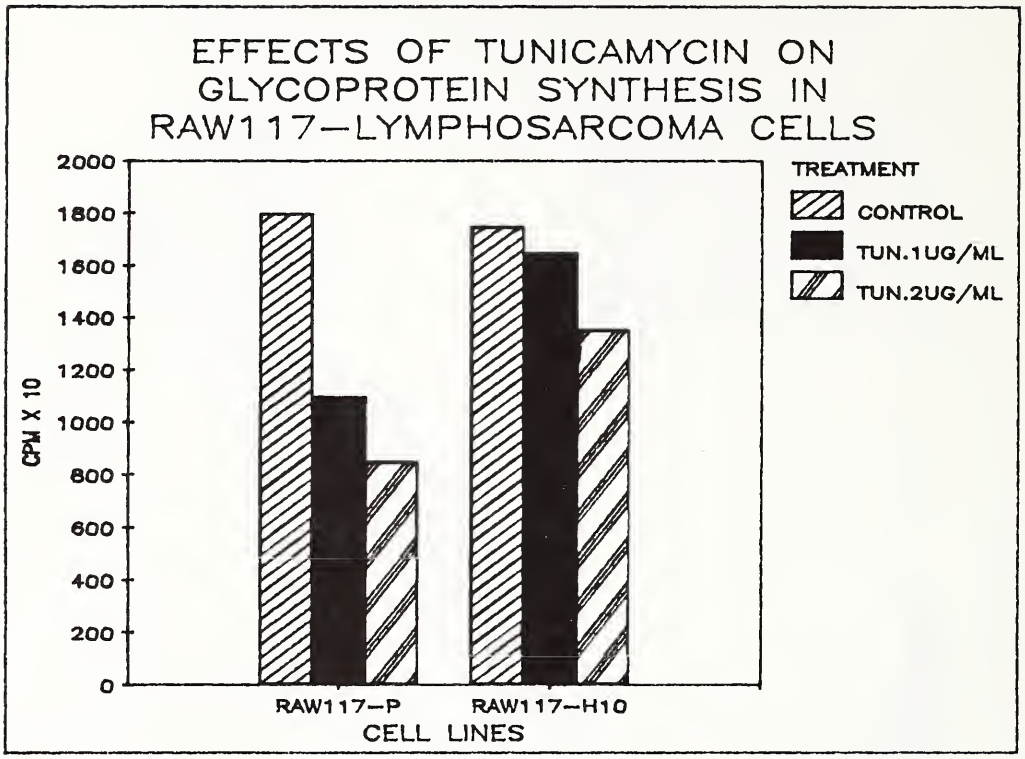

FIGURE 4: *5 x 10^3 cells/ml (5 ml/dish) were plated in 60mm petri dishes and incubated with or without appropriate concentrations of tunicamycin. Six hours before harvesting the cells, 0.5 μ Ci of ^{3}H-fucose was added. Each point represents the average of triplicate determinations of radioactivity (Cpm).*

phosarcoma cells. The lymphosarcoma cells grow in suspension culture, although a certain percentage of cells adhere to the culture dish. The percentage of adherent cell populations varies somewhat in cultures of both cell lines, and does not appear to be a constant feature of either the RAW 117-P or RAW 117-H10 cell line. Tunicamycin decreased the adherent cell population in both these cell lines. As shown in Table 1, in RAW 117-P cell line cultures the percentage of adherent cell population was decreased from 11% to 3% of the total cell population at a concentration of 1 μg/ml tunicamycin. A similar decrease in adherent cell populations in the RAW 117-H10 tumor cell cultures was noticed only at 5 μg/ml concentration of tunicamycin. At a tunicamycin level of 1 μg/ml, little or no decrease in adherence of RAW 117-H10 cells was seen.

TABLE 1. *Effect of Tunicamycin on Adherence Properties of Lymphosarcoma Cells*

CELL TYPE	% ADHERENT CELLS		
	Control	Tunicamycin 1 μg/ml	Concentration 5 μg/ml
RAW 117-P	11	3	N.D.
RAW 117-H10	19	17	7

N.D. = not done

Table I - 5 x 10^3 cells per ml were plated in 100mm x 15mm (1001 Falcon) petri dishes with or without (control) the appropriate amount of tunicamycin (1 μg/ml or 5 μg/ml). Cells were incubated for 36 hours at 37°C in an atmosphere containing 5% Co_2 and 95% air. Adherent cells were calculated then as described in the text.

Figure 5 depicts the *in vivo* properties of tunicamycin treated cells. Preliminary studies on the biological properties of tunicamycin treated cells revealed that in the experimental protocol used, tunicamycin increased the malignant properties of RAW 117-P cells but did not alter the malignant properties of RAW 117-H10 cells which are already highly malignant in nature. About seventy percent of the animals inoculated (6 mice per group) with tunicamycin treated (2 μg/ml) parental cells (RAW 117-P) died by day 26 post-injection but most control animals injected with untreated parental cells were all alive at that time. The effect of tunicamycin in increasing the malignancy of these cells appeared to be a dose dependent phenomenon, since tunicamycin treatment of RAW 117-P cells at a lesser concentration (l μg/ml) resulted in death of only 20% of the injected animals on day 26 and death of 50% of the injected animals on day 30.

Tunicamycin, a protein glycosylation inhibitor, is produced by *Streptomyces lysosuperficus* (12). It inhibits the growth of gram positive bacteria, fungi, yeast and viruses. Tunicamycin blocks the synthesis of N-acetylglucosaminyl pyrophosphoryl polyisoprenol by inhibiting the transfer of N-acetyl-glucosamine-1-phosphate to dolichol monophosphate (7). In most of the mammalian cells studied the amount of tunicamycin required for significant growth inhibition was at the sub-microgram level, usually 0.1 to 0.5 μg/ml (6, 7). A much higher concentration of tunicamycin was found to be required for growth inhibition of RAW 117-P and RAW 117-H10 lymphosarcoma cells. We observed a marked difference in the amount of tunicamycin required for growth inhibition of these two cell lines. The highly metastatic variant cell line RAW 117-H10 required almost 5 times more tunicamycin than the parental cell line RAW 117-P for similar growth inhibition.

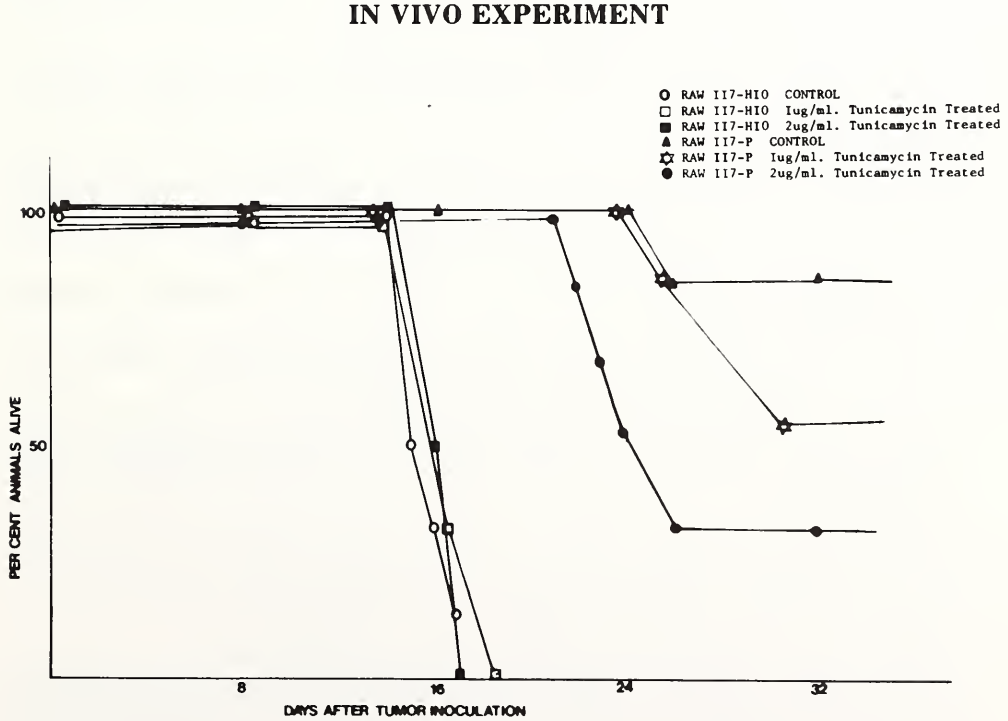

FIGURE 5: *5 x 10^3 viable tunicamycin treated or untreated RAW117-P or RAW117-H10 cells in a volume of 0.1 ml were injected into each mouse via the tail vein. Each group consisted of 6 animals. Death of the animals were noted daily.*

The reason for this increased resistance to tunicamycin by the highly malignant cell line is not known.

The expression of at least two major glycoprotein cell surface antigens (as recognized by xenoantiserum) "shifts" with altered malignant capacity of the RAW 117 lymphosarcoma cell lines. One of these antigens, a viral-coded glycoprotein (gp 70) is expressed much less on the highly malignant variant RAW 117-H10 cell line than on the less malignant parental RAW 117-P cells. It is likely that tunicamycin, through inhibition of glycosylation of cell surface components, prevents complete expression of major glycoprotein antigens such as gp 70, and may allow the less malignant RAW 117-P cells to masquerade as the more malignant RAW 117-H10 cells, perhaps at least long enough to establish experimental metastatic liver foci in the recipients. Quantitative assays of gp 70 and other cell surface glycoprotein expression will be necessary to see if the idea of molecular modulation of malignancy is correct. Further studies will also be required to determine the molecular basis for *in vitro* adherent difference in the tunicamycin treated and untreated cells. There appears to be no direct relationship between *in vitro* adherence and malignancy, since these adherence properties are similar in control (untreated) cultures of RAW 117-P and RAW 117-H10 cells.

Acknowledgments

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Membrane Redox Systems in Porcine Neutrophils

P. McLoughlin, I.L. Sun and F.L. Crane
Department of Biological Sciences
Purdue University
West Lafayette, Indiana 47907

Introduction

Neutrophils undergo a series of changes in their oxygen metabolism during phagocytosis or stimulation by a number of soluble compounds (3,15,24). These changes are referred to as the respiratory burst and are generally characterized by increases in oxygen consumption, superoxide and hydrogen peroxide production, and hexose monophosphate shunt activity. Activation of the respiratory burst is initiated by perturbation of the plasma membrane and does not require phagocytosis (21). The purpose of the burst appears to be (2,4,16) to provide the cell with a group of highly reactive products which the cell can use for the destruction of invading microorganisms. Many studies indicate that the respiratory burst results from (5,11,14) the activation of an enzyme in the plasma membrane which catalyzes the reduction of oxygen to superoxide using NADPH or NADH as the electron donor. The burst can be activated by a wide variety of agents. In the present study, concanavalin A, phorbol myristate acetate, and sodium fluoride were employed to stimulate the respiratory burst. In this study the aspect of the respiratory burst focused upon was the stimulation of superoxide production.

The existance of a redox enzyme which is transmembranous in orientation has been shown in the plasma membrane of both animal (7,9,13) and plant (8) cells. This enzyme transmits electrons from internal reducing compounds to the impermeable electron acceptor ferricyanide (22,17). NADH has been shown to be the electron donor (8,9) which is reducing external ferricyanide. Löw and Crane (18,19) have proposed that this enzyme may be involved in the control of cellular metabolic functions because it is affected by physiological levels of anabolic and catabolic hormones. This idea is further supported by the work of Sun and Crane (25) which shows this enzyme to be inhibited by several anticancer drugs.

The ability of the neutrophil to reduce oxygen to the superoxide anion has been well established. In this paper we examine both the superoxide production and the transmembranous dehydrogenase activities of porcine neutrophils. Superoxide dismutase sensitive cytochrome c reduction is often used as a detector of the superoxide anion (20). Ferricyanide reduction is used to measure the activity of the transmembranous dehydrogenases (7). By observing each systems response to three agents known to stimulate superoxide production, we attempted to determine if all membrane redox activity observed in porcine neutrophils is associated with the generation of superoxide or if there is some superoxide independent redox function at the surface of porcine neutrophils.

Materials and Methods

Horse heart ferricytochrome c (grade VI), superoxide dismutase Type 1: from bovine blood, 4-phorbol 12-myristate 13-acetate, concanavalin A (Type IV) and sodium fluoride were purchased from Sigma Chemical Co., St. Louis, Mo All other reagents were of analytical grade.

Porcine neutrophils were prepared from blood that was treated with 15 mM ethylenediaminetetraacetate (EDTA) to prevent coagulation. Neutrophils were

isolated using a modification of the procedure of Dioguardi et al. (12). Blood was centrifuged at 3000 g for 10 min., plasma was removed by aspiration and the neutrophil rich fraction was drawn off. This fraction was then treated with .83% NH_4Cl in a ratio of 1 part blood:8 parts NH_4Cl and centrifuged at 120 g for 2 min. The resulting pellet was washed with .83% NH_4Cl and centrifuged at 120 g for 20 min. A third wash was done to ensure elimination of contaminating erythrocytes and platelets, resulting in a cell suspension composed primarily of neutrophils. Neutrophils were suspended in whole cell buffer (pH 7.0 containing 137 mM NaCl, 2.7 mM KCl, 1.2 mM $CaCl_2$, 8.4 mM NaH_2PO_4, 7.3 mM sucrose, 0.5% dextrose and 2% fetal calf serum.

Neutrophil suspensions were treated with agents which stimulate superoxide generation and incubated at 37°C with constant stirring for 3 min. Cells were determined viable using Trypan blue exclusion. Incubation conditions varied with the stimulator. The assay buffer used for concanavalin A, and phorbol myristate acetate was whole cell buffer. The assay buffer for sodium fluoride was based on Dulbecco's phosphate saline (PBS) (10) and contained .117 mM NaCl and .31 mM $CaCl_2$. This low salt buffer was used to correct for the additional electrolytes from sodium fluoride and to prevent the precipitation of calcium fluoride. In assays with less than 20 mM sodium fluoride, NaCl was used to restore tonicity.

Cytochrome c and ferricyanide reduction were measured with an Aminco DW-2a spectrophotometer in the dual wavelength mode, subtracting absorbance changes at 541 nm from 550 nm for cytochrome c reduction and at 500 nm fom 420 nm for ferricyanide reduction. Extinction coefficients used for calculation of ferricyanide and cytochrome c reduction were 1 and 19 mM^{-1} cm^{-1} respectively.

The reaction mixture for ferricyanide reduction contained whole cell buffer, 0.3 mM potassium ferricyanide, the stimulator and superoxide dismutase concentrations as specified, in a total volume of 3.0 mls. The conditions for cytochrome c reduction were the same except 67 μg/ml cytochrome c and 0.33 mM potassium cyanide were substituted for ferricyanide. The average concentration of neutrophils in the final reaction mixture was 0.00953 g cell wet weight.

Results

Porcine neutrophils were assayed in the absence of a stimulus in order to obtain a resting or control rate for cytochrome c and ferricyanide reduction. The effect of superoxide dismutase (SOD) on these resting rates is shown in Fig. 1 and is expressed as percent inhibition of the respective control rate. The maximum inhibition of the resting rate obtained in the presence of SOD are 25% and 35% for ferricyanide and cytochrome c respectively.

We examined the effects of three agents, which are known to stimulate superoxide production, by monitoring the rates of cytochrome c and ferricyanide reduction. When phorbol myristate acetate (PMA) was the stimulus employed (Fig. 2), there was up to 500% stimulation above the control rate of cytochrome c reduction. In addition, PMA shows a 150% stimulation above the ferricyanide control rate. The effect of SOD on these PMA(15 ng/ml)-stimulated rates of reduction is shown in Fig. 3. When the rates of reduction were followed in the presence of SOD there was 90% maximum inhibition of the PMA-stimulated rate of cytochrome c reduction. PMA-stimulated ferricyanide reduction showed a 60% maximum inhibition in the presence of SOD.

Concanavalin A (Con A) showed a 5-15% stimulation in the rate of ferricyanide reduction and a 10-135% stimulation in the rate of cytochrome c reduction (Fig. 4). At high concentrations of Con A (2.5 mg/ml) there is a further increase in

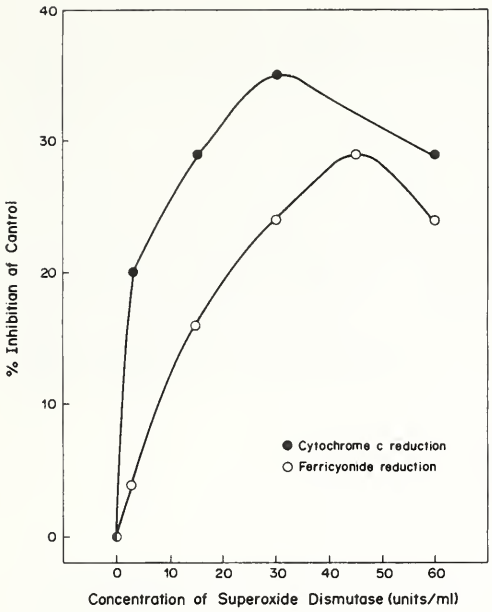
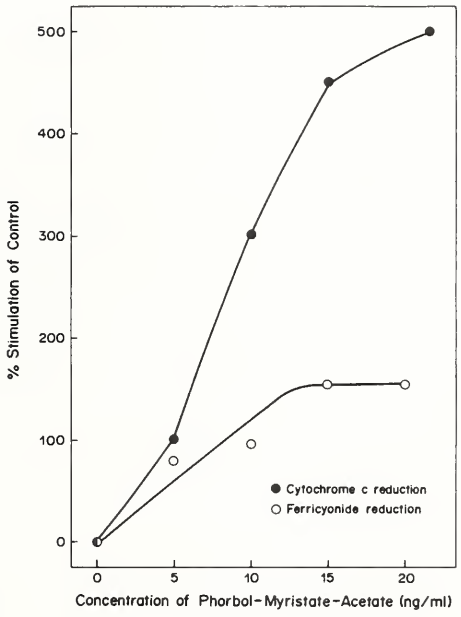


FIGURE 1. *Effect of superoxide dismutase on cytochrome c and ferricyanide reduction in porcine nuetrophils. Control rates of 16.19 and 218.2 nmol/min/g cells wet weight for cytochrome c and ferricyanide reduction respectively were obtained.*

FIGURE 2. *Effect of phorbol myristate acetate on cytochrome c and ferricyanide reduction in porcine neutrophils. Control values of 3.93 and 242.67 nmol/min/g cells wet weight were obtained for cytochrome c and ferricyanide reduction respectively.*



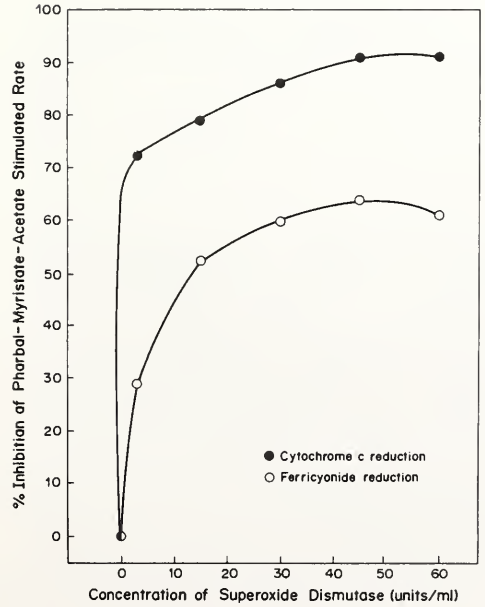


FIGURE 3. *Effect of superoxide dismutase on phorbol myrisate acetate (15 ng/ml) stimulated cytochrome c and ferricyanide reduction in porcine neutrophils. Control values of 25.68 and 487.46 nmol/min/g cells wet weight for cytochrome c and ferricyanide reduction respectively were obtained. The phorbol myristate acetate (15 ng/ml) stimulated rates for cytochrome c and ferricyanide reduction were 143.46 and 1285.63 nmol/min/g cells wet weight respectively.*

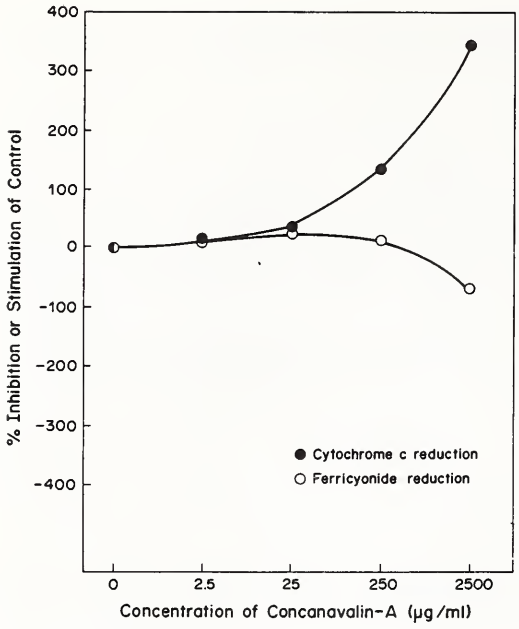
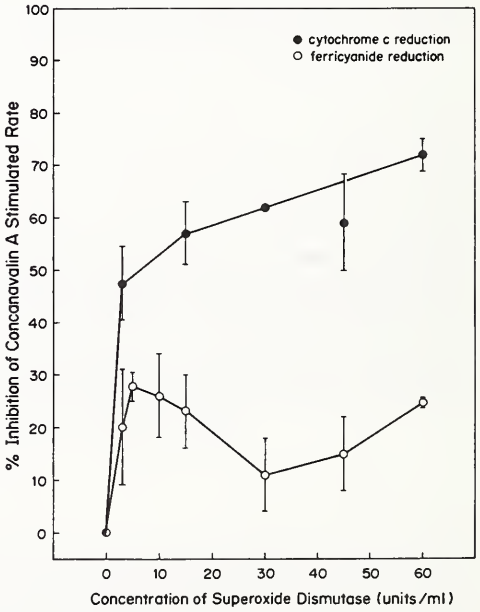


FIGURE 4. *Effect of concanavalin A on cytochrome c and ferricyanide reduction in porcine neutrophils. Control values of 16.38 and 136.05 nmol/min/g cells wet weight were obtained for cytochrome c and ferricyanide reduction respectively.*

FIGURE 5. *Effect of superoxide dismutase on concanavalin A (250 µg/ml) stimulated cytochrome c and ferricyanide reduction in porcine neutrophils. Control values of 1.15 and 355.19 nmol/min/g cells wet weight were observed for cytochrome c and ferricyanide reduction, respectively. The concanavalin A (250 µl/ml) stimulated rates of cytochrome c and ferricyanide reduction observed were 2.34 and 409.84 nmol/min/g cells wet weight respectively.*



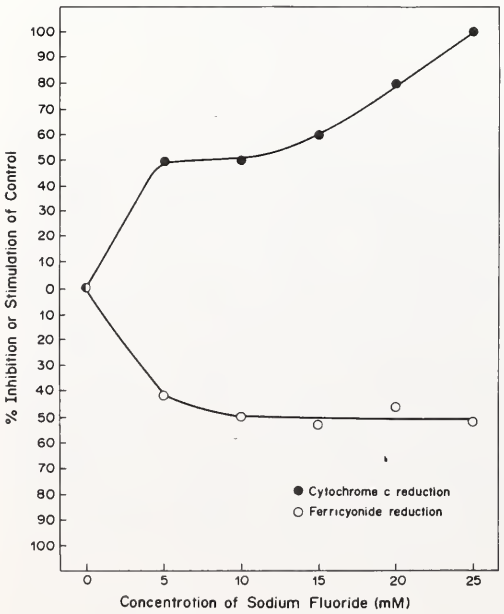


FIGURE 6. *Effect of sodium fluoride on cytochrome c and ferricyanide reduction in porcine neutrophils. Control values of 5.60 and 125.18 nmol/min/g cells wet weight were obtained for cytochrome c and ferricyanide reduction respectively.*

the cytochrome c rate, while the rate of ferricyanide reduction is inhibited. We examined the effect of SOD on the Con A (.25 mg/ml) stimulated rates of both ferricyanide and cytochrome c reduction as shown in Fig. 5. It was observed that a maximum of 75% of the Con A-stimulated cytochrome c rate is inhibited by SOD. The maximum inhibition of the ferricyanide rate achieved with SOD is 30% of the Con A-stimulated rate reduction.

The last agent used was sodium fluoride (NaF). At all concentrations of NaF employed in these experiments there was a stimulation in the rate of cytochrome c reduction and an inhibition in the rate of ferricyanide reduction, as shown in Fig. 6. The effect of SOD on the NaF (200 mM)-stimulated cytochrome c reduction is shown in Fig. 7. There was a maximum of 50% inhibition of this rate in the presence of SOD.

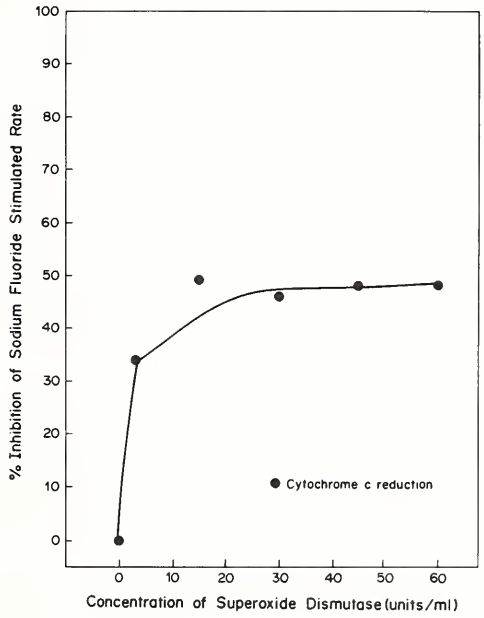


FIGURE 7. *Effect of superoxide dismutase on sodium fluoride (20 mM) stimulated cytochrome c reduction. Control value for cytochrome c reduction obtained was 28 nmol/min/g cells wet weight. The sodium fluoride (20 mM) stimulated rate of cytochrome c reduction was 47.45 nmol/min/g cells wet weight.*

Discussion

The results in this paper are in accord with earlier reports dealing wth the superoxide generating enzyme (3,15,16,24). The agents PMA, Con A, and NaF all increased the rate of cytochrome c reduction. A majority of this stimulated rate of cytochrome c reduction, 90% and 75% for PMA and Con A respectively, was found to be sensitive to SOD. These rates could therefore be attributed to the generation of superoxide. The results for NaF differ in that only 50% of the stimulated rate is SOD sensitive. This suggests that NaF may be stimulating some reduction of cytochrome c by a pathway which does not depend on superoxide. Our data indicates different modes of stimulation of neutrophil metabolism by various agents. The fact, that NaF activated system is different from that of Con A in human neutrophils, also suggests more than one mechanism exist for activation of the respiratory burst enzymes (21).

Evidence has shown that fluoride-mediated activation of the respiratory burst in human neutrophils requires the presence of Ca^{++} (10). Our assay buffer for NaF is based on Dulbecco's PBS which contains a considerable amount of Ca^{++} (0.31 mM). Higher NaF stimulated rate may be reached under a higher Ca^{++} concentration, however it can not be achieved due to a precipitation of CaF_2 in the incubation mixture.

The present work also looks at the transmembranous dehydrogenase that has been observed in several cell types (8.9). When ferricyanide reduction was examined, a different response than that observed with cytochrome c was noted for all stimuli employed. When NaF was the stimulus, opposite responses were observed for the two systems. When the rate of ferricyanide reduction was stimulated, as was the case with PMA and Con A, the SOD sensitivity was examined. It was observed that 30% and 60% of the stimulated rates for Con A and PMA respectively were inhibited in the presence of SOD, indicating that very little of the Con A-stimulated ferricyanide rate is the result of superoxide generation. It can also be seen that the PMA-stimulated rate of reduction can not be solely attributed to generation of the superoxide anion. The enzyme involved in the ferricyanide reduction does not seem to play a major role for superoxide production in the respiratory burst of neutrophils.

Evidence has been presented that the activated form of oxidase for the respiratory burst is on the plasma membrane of phagocytic cells (23). It has also been reported (1) that the main superoxide forming oxidase is embedded in the plasma membrane of human neutrophils with a portion extending into the cytoplasm and the rest buried in the lipid bilayer. Our data suggests there are at least two membrane redox systems in the plasma membrane of porcine neutrophils: 1) the redox system involved in the reduction of oxygen to superoxide, using NADPH or NADH as the electron donor, and 2) the transmembranous dehydrogenase which transmits electrons from internal reducing compounds to the impermeable electron acceptor ferricyanide.

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Changes in Exogenous Palmitic Acid Utilization and Lipid Composition in Aging Cultures of *Penicillium chrysogenum*

Mary Lee Richeson
Department of Biology
Indiana University-Purdue University at Fort Wayne
Fort Wayne, Indiana 46805
and
Alice S. Bennett
Department of Biology
Ball State University
Muncie, Indiana 47306

Introduction

Growth characteristics of submerged cultures of *Penicillium chrysogenum* have been related to a predictable change in pH of the culture medium (S. Zoghbi, Master's Thesis, Ball State University, 1975). When grown at an initial pH of 7.4, cultures remained at this pH for approximately 20-40 hours, depending on culture conditions. Then during the next 9 hours, the pH dropped to 3.4-3.8 where it stabilized. The greatest increase in dry weight (growth) of cells occurred during the nine hours of the pH drop after which time 60% of the initial energy source (glucose) still remained in the medium. This indicated that the cultures were not nutritionally depleted at that time, rather it was demonstrated that glucose was not depleted until approximately 20-30 hours later. With these criteria as bases, young and old cultures were defined in this study as those harvested before and after the decline in pH.

This research reports upon the morphological characteristics and the lipid composition of both young and aging cultures. Differences in incorporation and utilization of radioactively labeled palmitic acid by young and aging cultures were studied using biochemical and autoradiographical techniques.

Methods and Materials

Stock cultures of *Penicillium chrysogenum*, Q 176, were maintained on potato-dextrose-agar slants and stored at 4 C. Spores were scraped and washed from slants with 10 ml sterile medium, transferred to cuvettes, adjusted to 30% transmittance at 520 nm, added to 40 ml sterile medium of modified Czapek formula (1) and incubated at 28 C for 16 hr on a reciprocating shaker (180 oscillations/min). This starter culture was added to 450 ml of medium in a 2800 ml Fernbach flask and incubated as above. The pH of the submerged cultures was monitored at 2 to 3 hr intervals until the pH declined and stabilized. Cells were observed by phase microscopy at similar intervals. Young (pH 7.2, 20 hr) and aging (pH 3.4, 40 hr) cultures were labeled with 2 μCi of 1-(^{14}C)-palmitic acid which had been made soluble in 1% albumin in 30% ethanol. After 2, 10, 60, and 120 min, 20 ml samples of the cultures were removed, filtered through Miracloth, and washed with distilled water. A small portion of mycelium from each was placed in 3% gluteraldehyde and fixed for electron microscopy. The remainder of the harvested mycelium was placed in 10 ml of methanol and used in lipid analysis.

Lipid Analysis

Mycelial samples were sonified for two minutes by a series of 20 sec pulses

(setting 6, Branson Sonifier, Model J-17A, Heat Systems). Lipids from the ruptured cells were extracted with chloroform/methanol according to a modified Folch procedure (2). Total lipids were separated into lipid classes by thin layer chromatography on plates coated with silica gel G impregnated with 0.02% Rhodamine G using n-hexane/ether/glacial acetic acid/absolute methanol (90:2:2:3, by vol) followed by n-hexane/ether (94:6, by vol) (3). Bands were scraped into scintillation vials which contained 2,5-diphenyloxazole-1,4-bis (2-(5-phenyloxazolyl)-benzene (PPO-POPOP) in toluene and radioactivity was determined in a Beckman ambient temperature liquid scintillation system (LS150, Beckman Instruments, Inc.). To separate phospholipids, TLC plates (Adsorbosil-5 prekotes; Applied Science Div., Milton Roy Co.) were prewashed in n-hexane/ether (94:6 by vol) and dried. Total lipids were then applied and neutral lipids separated by Solvent System I, n-hexane/ether (94:6 by vol) followed by Solvent System II, chloroform/methanol/n-propanol/0.25% KCl/ethyl acetate (25:15:25:9:25 by vol) to separate the phospholipids (9). For visualization, plates were sprayed with Phospray (Supelco, Inc.). Separations were identified by comparision with standards scraped from plates and radioactivity determined.

Electron Microscope Autoradiography

Mycelia were fixed in 3% gluteraldehyde at 4 C for 2 hr, washed in buffer, post-fixed in 1% osmium tetroxide, and dehydrated for 5 min or less in each cold alcohol solution. By omitting a propylene oxide dehydration step the amount of incorporated label extracted by solvents was held to less than 7% (7). Mycelia were embedded in Epon 812 (Polyscience, Inc.) according to the procedure of Luft (5).

Ultra-thin sections (0.05 μm, silver or gold interference color) were cut with glass knives using 1KB Ultrotome (type 4801A, Ultramicrotome) and positioned on celloidin-coated slides according to a modified procedure of Kopriwa (4). Sections were stained with 1-2.5% aqueous uranyl acetate and Reynold's lead citrate, and carbon coated (5-10 nm). Slides were coated with Ilford L-4 nuclear tracking emulsion (Ilford Ltd., Ilford, Essex, England) and stored in light-sealed black boxes to await exposure. Following exposure they were developed in Kodak Formula D-170 (Eastman Kodak, Rochester, NY), fixed in sodium thiosulfate fixer and washed in running water. Within 15 min after developing, the celloidin film was floated free on a large tank of dust-free distilled water, 200 mesh EM grids were positioned over the sections on the floating film and the support film picked up on the ball of an acetone-cleaned evaporative flask and air dried. Grids were cut free, examined, and photographed using a RCA EMU-3C electron microscope.

Results and Discussion

Mycelia removed from rapidly growing submerged cultures showed changes in cell morphology with increased age (Figure 1). Young hyphae (harvested at 16 hr, pH 7.2) were slender with dense cytoplasm and infrequent end walls. This appearance contrasted sharply with the stubby thick-walled, highly vacuolar and granular appearance of aging cells (30 hr, pH 3.8). These changes do not appear to be caused by depletion of the energy source since most of the initial glucose remained in the medium at that time. Instead, observations suggest a change in utilization of products synthesized by the cell.

Thin layer chromatograms of extracted lipids indicated negligible amounts of triglycerides in young cultures whereas triglycerides were found in older cultures (Figure 2). There appeared, also, to be more diglycerides in young cultures.

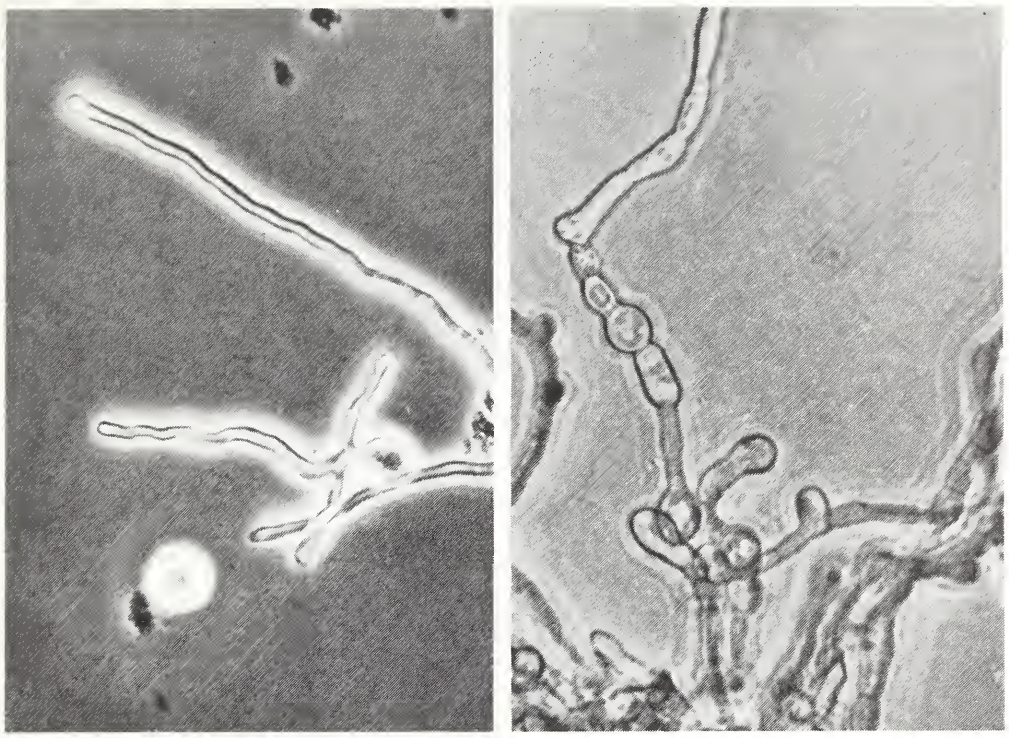

FIGURE 1. *Phase micrographs of mycelia of P. chrysogenum after (A) 16 hr. (pH 7.0) and (B) 40 hr. (pH 3.8) of growth. Magnification = X560*

However, the distribution of lipids does not reflect the metabolic activity of the cells as evidenced by the results of incorporation of labeled palmitic acid into young and older cultures.

Both young and old cultures incorporated large amounts of label (Table 1). Young cultures converted palmitic acid rapidly (within 2 min) into phospholipids.

TABLE 1. *Incorporation of Labeled Palmitic Acid Into Lipid Classes*[a]

Time[b]	Percent label[c] incorporated into:				
	PL	DG	FA	TG	CE
2 min					
Young[d] (2)	59.0 ± 4.8	9.9 ± 2.1	13.1 ± 1.7	11.9 ± 0.2	5.8 ± 0.7
Aging (5)	15.5 ± 2.7	6.1 ± 0.9	71.9 ± 1.3	5.0 ± 1.5	1.3 ± 0.6
10 min					
Young (2)	55.4 ± 0	8.7 ± 0.5	14.8 ± 2.6	14.7 ± 3.4	6.3 ± 0.3
Aging (5)	23.8 ± 1.5	6.6 ± 0.8	63.5 ± 3.0	4.0 ± 0.5	1.9 ± 0.6
60 min					
Young (2)	72.5 ± 1.2	4.0 ± 0.0	10.6 ± 3.3	10.9 ± 3.3	2.0 ± 0
Aging (5)	23.0 ± 4.4	5.7 ± 0.9	68.0 ± 6.0	4.2 ± 0.7	1.3 ± 0.8
120 min					
Young (2)	80.2 ± 0.4	3.1 ± 0.1	14.7 ± 0.3	1.7 ± 0.5	1.0 ± 0.3
Aging (5)	24.8 ± 6.5	5.7 ± 2.3	65.0 ± 7.8	3.3 ± 0.3	1.0 ± 0.3

[a]The value represents the mean of the number of experiments indicated in parentheses + S.E. Results are expressed in percent of label incorporated into lipid classes at the times indicated. Of the 218.4 nmol (2uCi) of label in the form of 1-(^{14}C)-palmitic acid added to each culture, young cultures incorporated 21.84 nmol after 120 min; aging cultures incorporated approximately 2 times this amount after 120 min incubation. Values for young cultures represent the mean of 2 samples, for aging cultures the mean of 5 samples. [b]Time after addition of label. [c]From 900 to 85,000 cpm recovered from individual cultures. [d]Young harvested at pH 7.2; aging, pH 3.5, PL – phospholipids, DG – diglycerides, FA – free fatty acids, TG – triglycerides, CE – sterol esters.

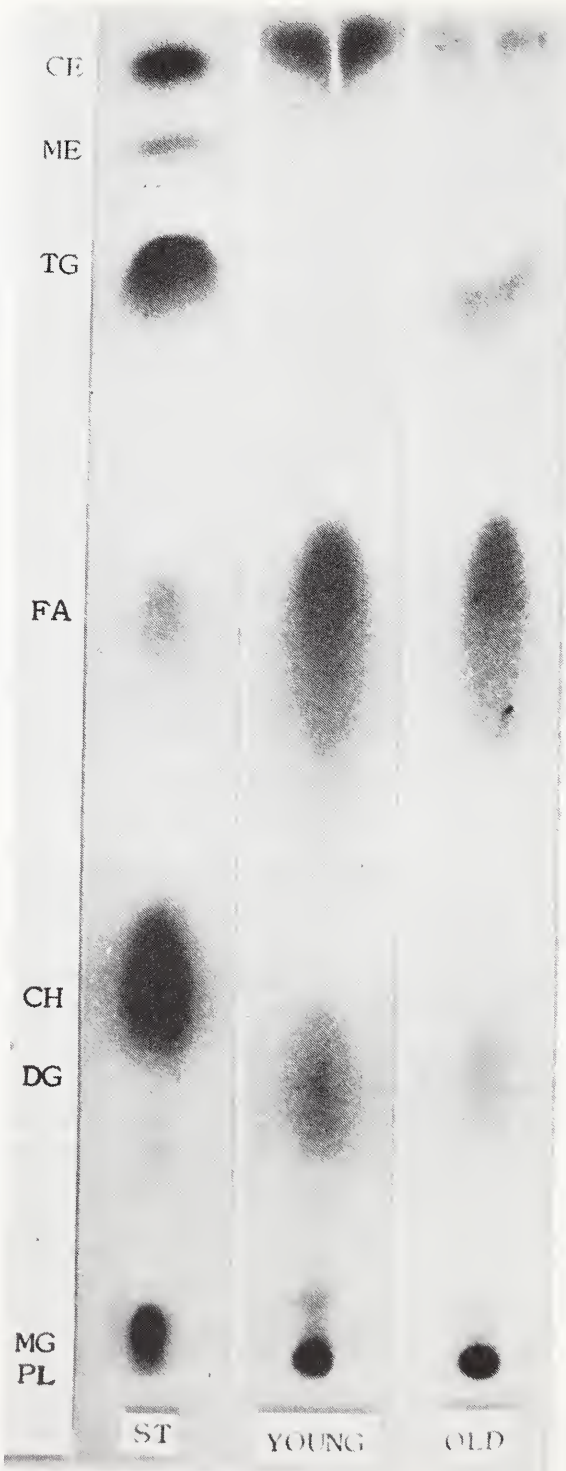

FIGURE 2. *Distribution of Lipids from Young and Old Cultures of P. chrysogenum. Spots were visualized by spraying with conc.* H_2SO_4 *and heating for 10 min. in an oven at 100°C.*
Standards top to bottom:
Ce—cholesterol (sterol) esters, ME—methylesters, TG—triglycerides, FA—free fatty acids, CH—cholesterol (sterols), DG—diglycerides, MG—monoglycerides, PL—phospholipids

Largest amount of label was recovered in the phospholipid fraction at all time points; less than 15% of the label remained in the free fatty acid fraction in young cultures. The conversion of the palmitic acid was slow in old cultures and less was converted into phospholipids. In aging cultures 30% of the labeled fatty acid was incorporated into complex lipids. Approximately 65% of the label remained in the free fatty acid fraction.

Incorporation of labeled palmitic acid into diglycerides, triglycerides, and

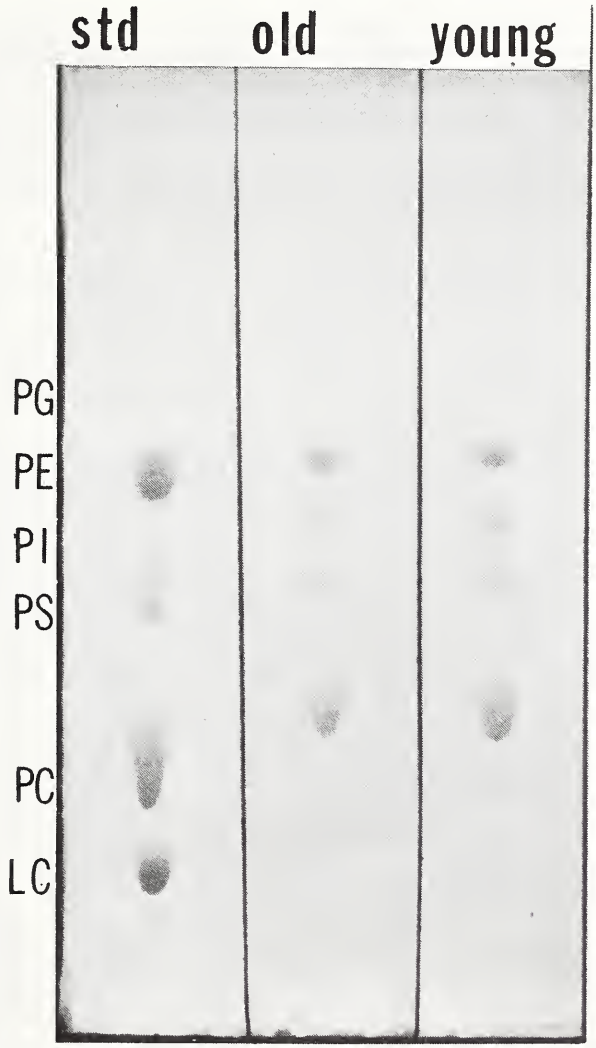

FIGURE 3. *Phospholipids of Penicillium chrysogenum. TLC plates were prewashed in n-hexane/ether (94:6 by vol) and placed in 100° C drying oven for 30 minutes. Total lipids were applied and neutral lipids separated by Solvent System I, n-hexane/ether (94:6 by vol) followed by Solvent System II, chloroform/methanol/n-propanol/0.25% KCl/ethyl acetate (25:15:25:9:25 by vol) to separate phospholipids. Visualization was by acid molybdate.*

Standards: LC—lysophosphatidyl choline
PC—phosphatidyl choline
PS—phosphatidyl serine
PI—phosphatidyl inositol
PE—phosphatidyl ethanolamine

The spot above PE was identified in other separations to be phosphatidyl gylcerol—PG.

sterols was similar in both culture conditions. Diglycerides, triglycerides, and sterols incorporated some label at early time points, but the amount decreased with time suggesting that these substances were not being stored as such in the cells but were being depleted as they were incorporated steadily into synthesizing membranes.

Phospholipid types have not been previously described for *Penicillium chrysogenum*. Five types were identified: phosphatidyl serine, phosphatidyl inositol, phosphatidyl choline, phospatidyl ethanolamine, and phosphatidyl glycerol (Fig. 3). Small amounts of cardiolipin were also detected. These findings are consistent with analysis of *Cephalosporium* (8) and for a wide variety of fungi (10). Types of phospholipids found in young and aging cultures are similar, however distribution of ^{14}C-label in phospholipids differed (Fig. 4). The phosphatidyl serine component incorporated most label in aging cultures, while young cultures incorporated the most label in phosphatidyl choline. Aging cultures incorporated about

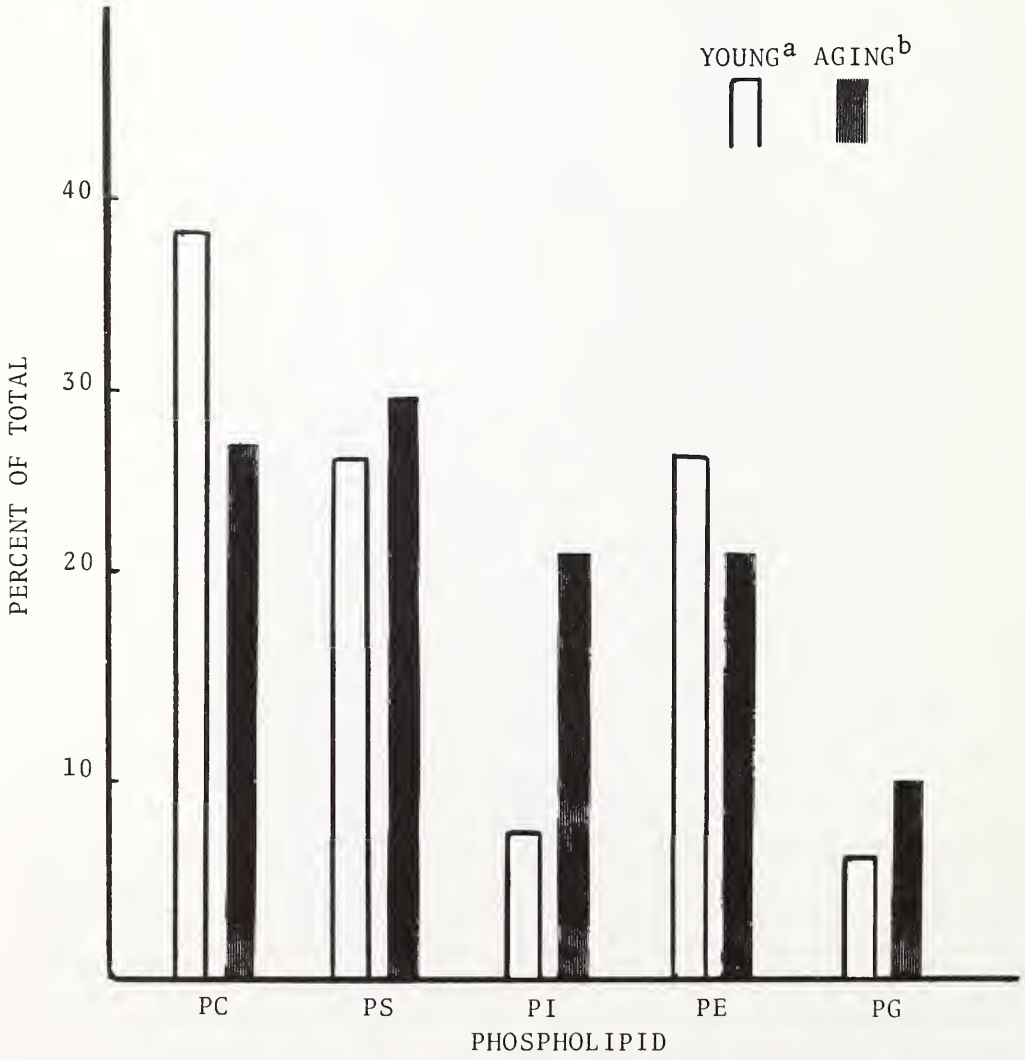

FIGURE 4. *Distribution of label in phospholipids of young cultures and aging cultures of P. chrysogenum incubated for 30 minutes with 1-(^{14}C)-palmitic acid. [a]Average of two samples, [b]average of five samples. PC-phosphatidyl choline, PS-phosphatidyl serine, PI-phosphatidyl inositol, PE-phosphatidyl ethanolamine, PG-phosphatidyl glycerol.*

TABLE 2. *Localization of 1-(^{14}C)-Palmitic Acid in Electron Microscope Autoradiographs of P. Chrysogenum*

Location of grains	Present[a] Young cells	Aging cells
Plasma membrane	69	47
Nuclear membrane	3	1
Cytoplasm (general)	26	26
Spherosomes	1	11
Mitochondria	1	15

[a]Number given is the percent of total grains counted; for young cells, from a composite tally of 12 micrographs, for aging cells, a composite tally from 24 micrographs.

20% of label in phosphatidyl inositol, young cultures averaged less than 10% of the label in this fraction. The amount of label in phosphatidyl glycerol varied considerably in samples, with up to 10% seen in many old cultures, but with only 1-2% recovered from young cultures.

High resolution autoradiographs of cells incubated with 1-(^{14}C)-palmitic acid support the biochemical evidence pertaining to the localization of label. In young cells approximately 70% of the developed grains were found in close association with plasma membranes while in aging cells no more than 47% of the developed grains were over the plasma membranes (Table II, Figure 5). In both young and

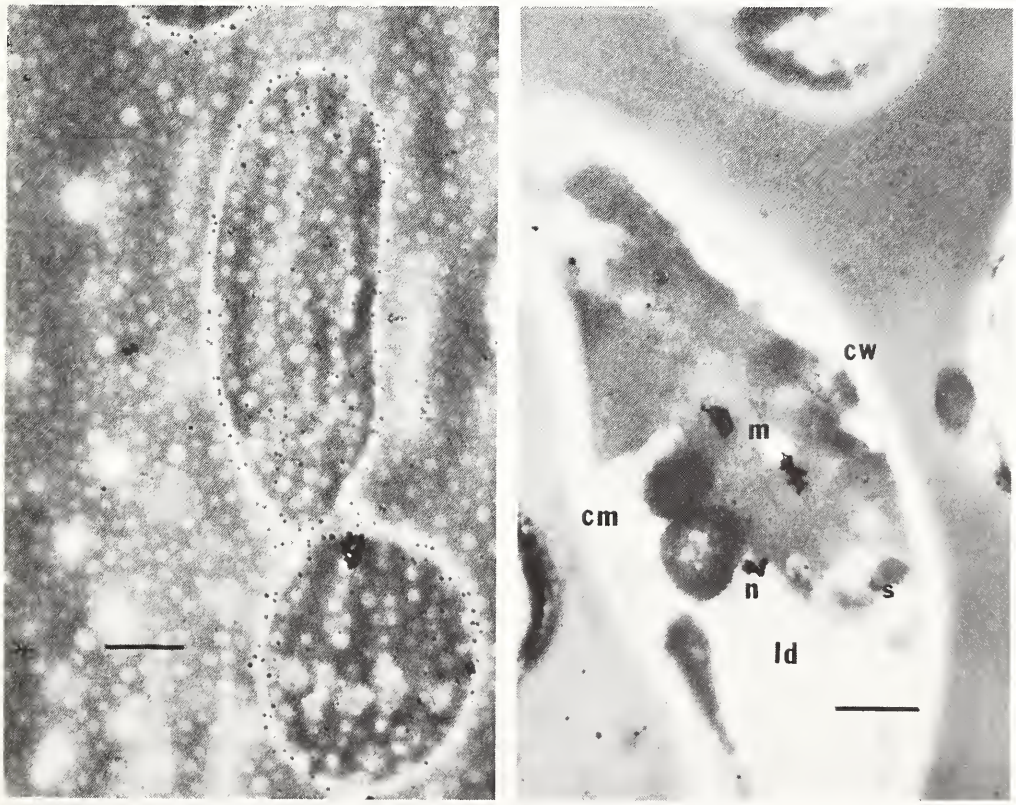

FIGURE 5. *Electron microscope autoradiographs of P. chrysogenum. Young cells (A) were incubated for two minutes and aging cells (B) were labeled for 60 minutes with 1-(^{14}C)-palmitic acid. Small punctate grains show generalized distribution of incorporated labels in areas of membranes in young cells. In older cells, grains were highly concentrated in localized lipid deposits.*
cm—cell membrane, cw—cell wall, ld—lipid depot, m—mitochondrion, n—nucleus, s—spherosome. Bar = 1μm

old cells, 26% of the developed grains appeared throughout the cytoplasm. Spherosomes, cell organelles identified as "lipid bodies", but of uncertain function in fungi (11,6) were evident in all cells. However, since only 1% of developed grains was found in spherosomes in young cells, with approximately 10% in aging cells, it appears that spherosomes are not involved in lipid utilization in *Penicillium.*

These results indicate a relationship between age of the culture and utilization of an exogenous source of fatty acid. Old cultures exhibited a decreased ability to incorporate the added palmitic acid into phospholipids. The types of lipids present for synthesis of structural components play a significant role in forming the morphological characteristics of cells. Thus changes in amounts and types of lipids would be reflected in altered cell morphology. Therefore, the age of the culture should be carefully considered in studies involving morphology and lipid metabolism in organisms such as *Penicillium chrysogenum.*

Acknowledgments

This work was in part supported by a CORE grant from the College of Sciences and Humanities, Ball State University.

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PHYSICS AND ASTRONOMY

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Department of Physics
Taylor University, Upland, Indiana 46989

Chairman-Elect: Malcolm Hults
Department of Physics and Astronomy
Ball State University, Muncie, Indiana 47306

ABSTRACTS

Nonisothermal, Convecting Liquid States. Marshall P. Cady Jr., Department of Natural Sciences, Indiana University Southeast, New Albany, Indiana 47150.—— A horizontal parallel plate glass cell sandwiched between two heat baths has been used to evaluate the critical Rayleigh numbers for transitions between discrete convection states. The convection states are established by heating the water containing cell with the lower of the heat baths and cooling with the upper heat bath. This results in a liquid layer which is unstable to convective motion, provided the resulting buoyant force exceeds the viscous dissipation force. The parameter describing the relative magnitudes of these forces is the dimensionless Rayleigh number defined by

$$R = (g \propto d^2 C L^3 / k \eta) \Delta T$$

where L is the cell height (1.440 cm) and ΔT is the temperature difference between the upper and lower cell boundaries. A dual Savart plate interferometer with a 632.8 nm helium-neon laser light source is used to monitor both vertical temperature distributions within the cell and fluid motion.

The first, third, and fourth critical Rayleigh numbers have been successfully determined by analyzing the interferometer fringe patterns for R values between 9,200 and 720,000 at room temperature. Since R_I characterizes the transition fron nonconvection to the simplest time-independent convection, it was determined by evaluating the intercept of the observed temperature gradient at cell center with the gradient expected for pure heat conduction only. We find that

$$R_I = 1667 \pm 137.$$

This is in good agreement with the theoretical value of 1708.

R_{III} characterizes the transition from steady state convection to time-dependent convection. However, the amplitude of the time dependency of the fringe proved to be too small to analyze with a photocell so a sudden increase in the standard deviation of the temperature gradient with R was used to evaluate R_{III}. It is found that

$$R_{III} = 3.30 \times 10^4 \pm 12\%.$$

The transition R_{IV} from low amplitude time-dependent convection to high amplitude time-dependent convection was determined by photocell evaluation of the rate of fringe fluctuations verses R and extrapolation to a rate equal to zero. We find that

$$R_{IV} = 4.42 \times 10^4 \pm 14\%.$$

It proved to be impossible to evaluate the transition from two-dimensional steady convection to three-dimensional steady convection, R_{II}, with a Bryngdahl

interferometer. Furthermore, the transition from time-dependent to turbulent convection was coarsely evaluated by visual inspection of fringe as $R_V = 5x10^5$.

Viscosity Determination of Newtonian and Non-Newtonian Fluids. ROBERT H. L. HOWE, Eli Lilly Company, Lafayette, Indiana 47906, and DAVID J. HOWE, Esmark Corporation, Columbia, Missouri 65201.—The procedure for determining the viscosity of a heavy bio-oxidation substrate is presented. The mathematical relationship of the viscosity and the rotational speed is derived. Some experimental data are illustrated.

A Laboratory Study of Cosmic Ray Produced Radionuclides with Muons

JOSEPH L. BREEDEN
Martinsville High School
Martinsville, Indiana 46151

Muons are a major constituent of the secondary cosmic radiation detected at sea level. They comprise 63% of the secondary radiation while 21% are neutrons, 15% are electrons, and <1% are pions and protons. This secondary cosmic radiation is responsible for 30 - 40% of the total natural background radiation (NCRP Report No. 45).

Electrons, taus, and their respective neutrinos and antiparticles are members of the family of leptons along with the muon. Leptons are particles which interact exclusively through weak and electromagnetic forces. The muon possesses a mass of 105.7 MeV/c^2, charge of ± 1, and a spin of ½. It is simply a more massive version of the electron.

Both positive and negative muons are produced in the atmosphere via the decays: $\pi + \rightarrow \mu + + \nu\mu$ and $\pi - \rightarrow \mu - + \bar{\nu}\mu$.
These muons can then free decay by the processes:

$$\mu + \rightarrow e + + \nu e + \bar{\nu}\mu \text{ and } \mu - \rightarrow e - + \bar{\nu}e + \nu\mu.$$

The negative muons also can decay via a capture decay: $\mu^- + p \rightarrow n + \nu\mu$ with the proton being bound in the nucleus of an atom. In the capture decay, the muon first enters an orbit around the nucleus. It then falls rapidly (10^{-13} sec.) into the nucleus and interacts with a proton forming a neutron and a neutrino thus changing atomic number of the element. Muon decay differs from that of electron capture because the muon capture process deposits an average of 35 MeV to the residual nucleus and is thus capable of ejecting several neutrons from the atomic nucleus. Therefore, the atomic mass can be changed through muon capture decay as well as the atomic number. The rate at which this occurs is dependent upon the Z or atomic number of the element.

The process of greatest interest here is the muon capture decay. It was the intent of this experiment to obtain data through radiochemical analysis of muon interaction products. Information such as this is vital to more fully understanding interactions mediated by the weak force.

One approach at acquiring this data is to use a scintillation detector with appropriate electronics to mesure Δt, the time interval between the muon entering the detector and the muon decay. This detector is a new type employing a plastic box filled with water and coupled to a photomultiplier tube. The water has proven to be as good, if not better, as a scintillator compared to more expensive systems. Since water is almost a universal solvent, a chemical solution may be placed in the box allowing for great experimental flexibility at a low cost. A chemical of the desired Z was dissolved therein. This study yields new information on the muon capture decay lifetime in various elements.

For the scintillation detector, a simple electronics system was employed. As a muon enters, it emits Cêrenkov radiation which is picked up by a photomultiplier as a start signal. Shortly thereafter, a stop signal follows from the muon decay. These signals go through a Timing Filter Amplifier, a Constant Fraction Discriminator, a Time to Pulse Height Converter, and then into a Multichannel Analyzer. The data was graphed in the form of counts vs. Δt (Figure 1). The results of the present study show an average lifetime of 2.27 $\pm$ 0.17 μs

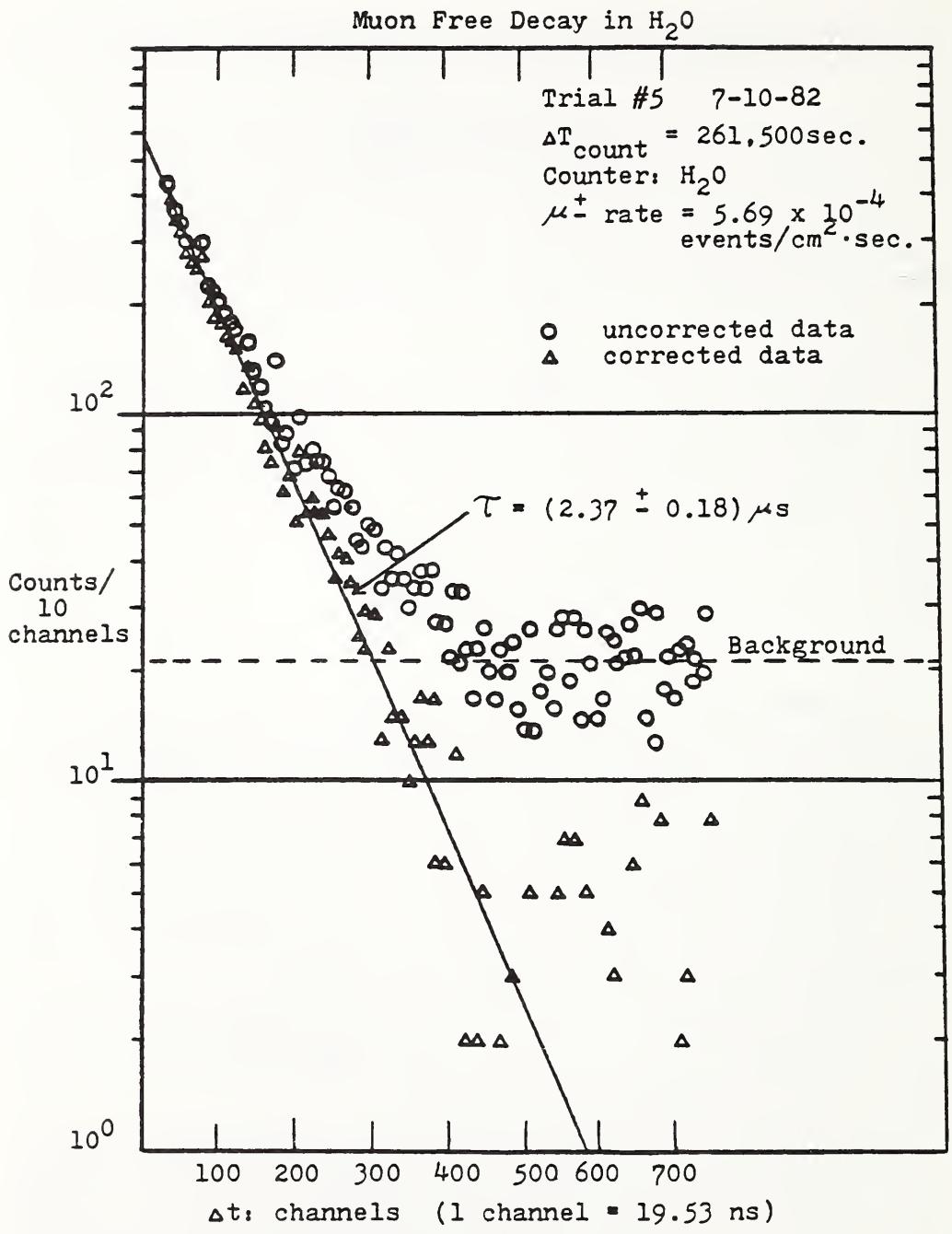

FIGURE 1

which is to be compared with the accepted free lifetime value of 2.19712 ± 0.000077 μs (Table 1), (Reviews of Modern Physics, 1980).

A second approach is to measure the rate of muon induced radioisotope production through a radiochemical analysis. The desired chemical was dissolved into a large volume of water, approximately 50-100 L to accommodate the low cosmic ray muon flux. Cadmium nitrate was used in order to produce Ag isotopes. Then $AgNO_3$ and HCl were added providing AgCl precipitate which was then filtered out. Rainwater was alternatively used since the ~1% concentration of

TABLE 1. *Muon Free Decay in H_2O Scintillation Detector*

Trial #	Total Events	Mean Lifetime	Counts/cm^2.sec.
3	2327	2.10 ± 0.07 μs	5.809 x 10^{-4}
4	1383	2.34 ± 0.44 μs	3.684 x 10^{-4}
5	6954	2.37 ± 0.09 μs	5.688 x 10^{-4}

argon in the atmosphere yields chlorine isotopes upon μ^- capture. The same chemical procedure was then followed. The gamma rays produced from the decay of the isotopes were then measured. In this manner, the distribution of the muon capture products could be measured.

In the large volume radiochemical analysis, entering muons preferrentially seek out the element of highest Z, due to charge transfer processes, and capture upon it. By utilizing a $Cd(NO_3)_2$ solution it is expected that ^{111}Ag, ^{112}Ag, ^{113}Ag, and ^{115}Ag are isotopes produced which can be detected using a standard Ge(Li) gamma ray detector. An energetic muon capture should release excess energy thus knocking neutrons out of the residual nucleus and allowing for the various silver isotopes. The distinctive gamma ray peaks of these isotopes were as follows: ^{111}Ag at 342 keV, ^{112}Ag at 617 keV, ^{113}Ag at 298 keV, and ^{115}Ag at 230 keV. Preliminary analysis has yielded negligible results due to high background and low count rates. Further experiments will be carried out in order to reduce the background and obtain a definite signal.

When using rainwater as a source of a sample, atmospheric Ar is converted into ^{34}Cl, ^{38}Cl, ^{39}Cl, ^{40}Cl as a result of the capture decay with subsequent neutron emission. The gamma ray peaks of these radionuclides are: ^{34}Cl at 146 and 2127 keV; ^{38}Cl at 1642 and 2168 keV; ^{39}Cl at 250.3, 1267, and 1517.4 keV; and ^{40}Cl at 1461 keV. Results of the rainwater study for the production of ^{39}Cl were plotted and compared with a previous study (Figure 2), (Young, et al., 1968). The first trial was a rainfall with a rate of 1.25 in./hr. which yielded 5 dpm/L. The second sample was obtained from a low rainfall rate of 0.025 in./hr. and yielded 70 dpm/L. These findings are in good agreement with Young, et. al. and indicate the general nature of rain formation from water vapor condensation on aerosol particles in the atmosphere. Young, et. al. have determined that the radionuclide concentration present in the aerosol particle depends upon the size of the raindrop formed. Therefore, the smaller the raindrop, the larger the dpm/L and the lower the rainfall rate. The log normal concentration of activity vs. the rainfall rate is simply inversely proportional to the rainfall rate or the size of the raindrops (Figure 2).

This project is part of a continuing study of muon interactions investigating the bound state quark structure of the nucleus. The cosmic ray muon data obtained from the scintillator measurements exhibit a suitable degree of accuracy at a greatly reduced expense. As a result of this study, an interesting discovery was made pertaining to the concentration of radioisotopes and the size of a raindrop. This may prove to be a valuable source of information concerning aerosol particle formation.

I wish to acknowledge the research advice and encouragement of Dr. Thomas Ward of IUCF and Mr. Walter Cory of the High School Science Student Institute. This work was performed under the auspices of the National Science Foundation.

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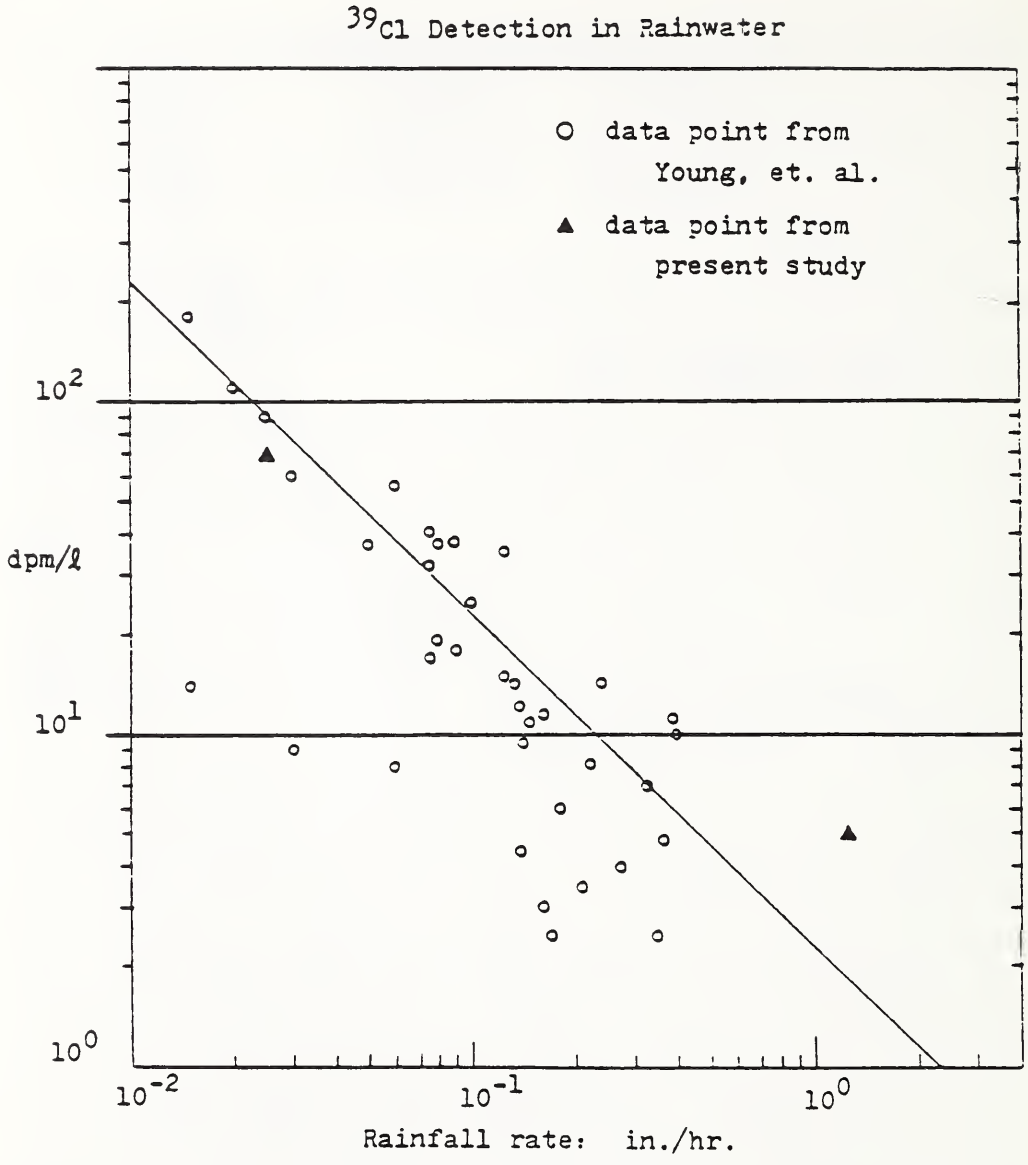

FIGURE 2

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Direct 3-D Measurement of High Energy Elementary Particle Interactions in Nuclear Emulsion Targets

GERALD P. THOMAS
Department of Physics and Astronomy
Ball State University
Muncie, Indiana 47306

Introduction

This work is an outgrowth of work carried out earlier by Thomas (1) which involved a partial automation via micro and macro computers of microscope measurements in nuclear emulsion target pellicles which have been previously exposed to high energy accelerator beams of primary projectile particles at large accelerator installations. The motivation was to reduce the labor in performing measurements as well as provide real-time measurements, calculations, and on-line re-measurement decision making. However, the earlier work suffered from several limitations which have been substantially eliminated here:

1) The Koristka research measurement microscope and computerized automation system was centered upon automation of just one axis; the single optical encoder was attached to just the y-axis. This was sufficient for so-called "multiple scattering" measurements, in which work for the x-y plane (projection plane) is sufficient for comparison with theory. But other types of measurement algorithms such as measurement of primary beam particle deflection angles (described below and due to sub-nuclear interactions) involve the depth (z) dimension as well. Yet, both of these latter algorithms involve only fixed x-axis coordinate separations (fixed cell lengths) between measurement points and so no automation of x-measurements is as greatly desirable. However, the system described herein allows for complete 3-d (x,y,z) measurement flexibility for arbitrary measurement algorithms within the general scope of the emulsion technique.

2) The counting of the square wave pulses outputted by the optical encoders was done by the micromcomputer (KIM-1) itself. This led to only marginally good pulse count totals when the y-axis microfilometer screw and attached scale (1) was rotated too rapidly, due to the finite number of machine level program steps needed to detect, read in, and store each pulse. The digital hardware counter system described below was specially constructed to alleviate this problem.

3) The macrocomputer (DEC-10 university computer) was a necessary part of the automation architecture: Once the KIM microcomputer system counted up the pulses whose number is proportional to the y (crosshair) motion per x (cell) change, and stored this number temporarily, it then had to dump its stored data block of count totals and demographic parameters for the event to a temporary DEC-10 data file which had already been opened up from the TTY over the modem just prior to the dump. A manual make-before-break switch was needed to go to full duplex TTY mode such that calculations, using a DEC-10 program, could be made and sent to the TTY for possible re-measurement prompting. All this was subject to university computer downtimes not to mention the inconvenience of setting up files that needed to be run for each event's measurement. To circumvent this, more RAM was obtained so that measurement and calculational programs in BASIC could be run on the KIM. Also an HDE dual floppy disk system was obtained so as to load programs and store output data for future reference, collations and graphing, and comparison with theory predictions.

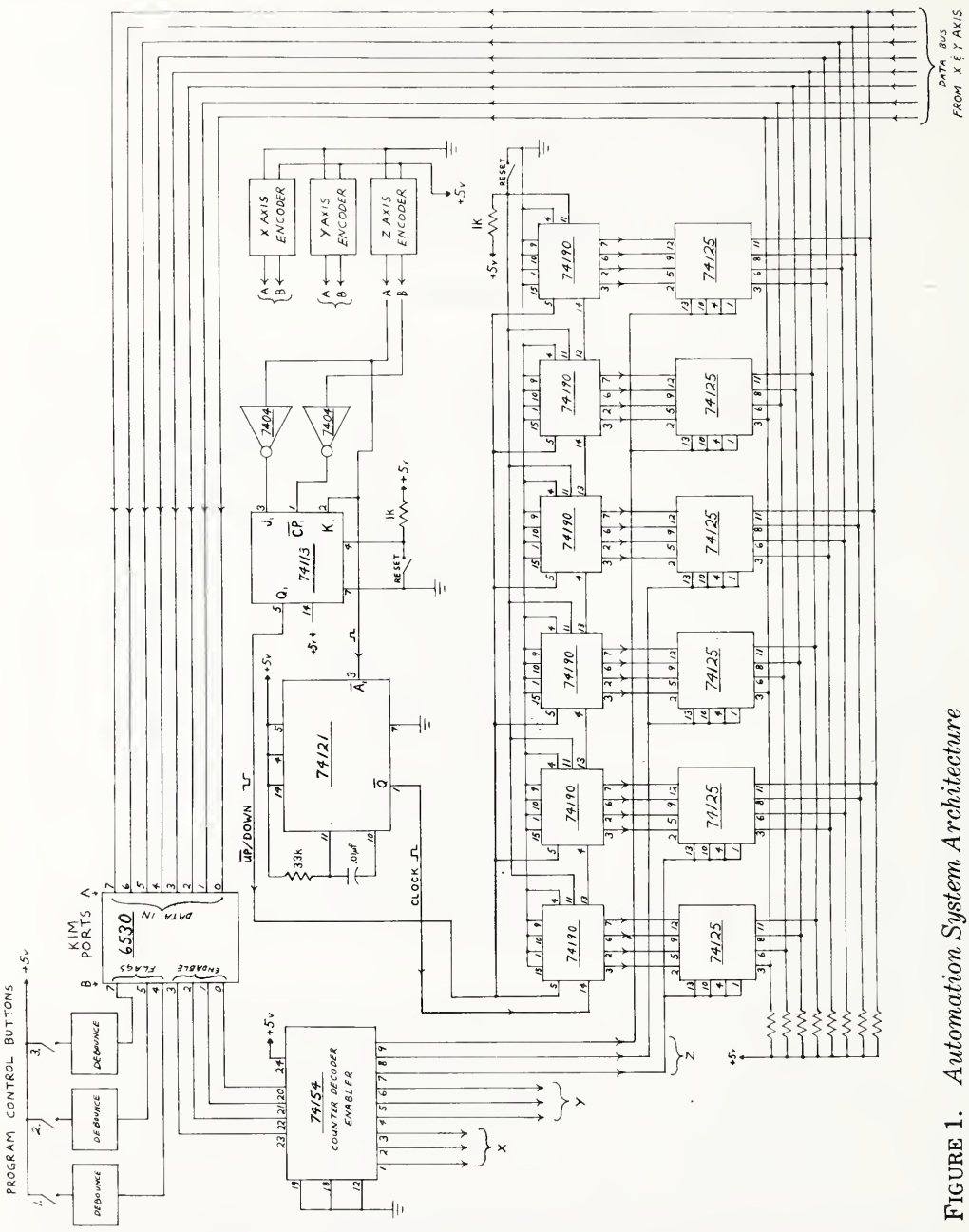

FIGURE 1. *Automation System Architecture*

Method

A schematic of the 3-d automation hardware architecture used is seen in Figure 1. More on-line hardware is used than cited before by Thomas (1); namely, an external digital counter module as well as two more optical encoders (for the x and z axis, although the angle measurement algorithm described below only reads the y and z axis encoders). Conventionally, the y-axis measurement uses an eyepiece microfilometer scale described in (1) which gives the y displacements that are needed in multiple scattering theory. The change of depth (z), as seen in the eyepiece, is measured by reading the z-collar scale; whereas the cell lengths between x-coordinate positions for various y,z data pairs are given on the x-collar scale or more simply by the audible "clicks" of a notched sleeve for definite lengths

of ex. 50, 100 microns, etc. The three A to D converters, namely the 3 optical encoders, are attached concentrically to the 3 microscope shafts. One whole detailed function is shown for one of the 3 duplicate axis systems, viz., the z-axis. Each encoder will supply a staggered pair of square wave pulses A and B (A leading the B train, if the collar is turned clockwise, looking into the scope, and visa versa) to the angular direction discriminator chips (74113 and 74121).

One function of the 74121 chip component of the direction discriminator is to pass only the first of the 2 pulse trains (A or B) from the encoder to the clock input of the up-down BCD digital counters (six U/D 74190 counter chip exist for each of the 3 axes). The other of the 2 outputs from the discriminator section enables (by using the 74113 chip) either the up or the down function of the counters. The capacitor and resistor connected to the 74121 are tuned to insure a small enough surviving pulse width such that the largest practical revolution speeds of the shaft encoder will not cause any pulse to be missed by the counter. Also, since the rear end of the leading pulse triggers the release of a pulse from $\overline{Q}$ of the 74121, the requirement that any changes in the up/down status pulse take place before the clock pulse from the 74121 reaches the 74190's is met such that no pulses are lost during changeover. On each axis board there are thus mounted six 74190 counters in series to get the six nibbles (3 bytes) of resolution per axis sought after (up to decimal 999,999) in this application. Each of the 6 decimal nibbles pass to corresponding 74125 tri-state buffers on the same board which are in turn connected by the 8-bit data bus to Port A of the KIM microcomputer. These 6 latched nibbles per axis are gated out to the bus, 2 nibbles(1 byte) at a time (Lo, then Mi, and finally Hi), by means of a series of 3 consecutive enable pulses (06,07, and 08 for the example of the z axis case) sent to the buffers by the MANG machine language subroutine of the calculation program used for the angle algorithm here and written in BASIC called RANG. In particular, for each enable nibble from bits 0-3 of the KIM control of Port B, the 74154 decoder chip produces, for each axis, the correct set of 3 gate pulse enables to get the data byte from each pair of the six 74125 tri-state buffer latches. Both MANG and RANG are stored on the systems disk and the disk data file MUGET on the applications disk as shown in Table 2. Machine level files are designated in the HDE/KIM DOS system by % and programs in BASIC by @.

The machine program (MANG) is shown in detail in Table 2. In the angle measurement algorithm described here, the machine subprogram MANG, written in the 6502 machine language program set (see the left side of Table 2), communicates with the "FOR-NEXT" data INPUT loop of the aforementioned BASIC computation program RANG via a flag system in which one of 3 possible flags are set (using 3 de-bounced button switches next to the KORISTKA microscope, see Figure 1). In particular, one or another of the series of 3 flag buttons next to the scope is depressed at appropriate times during an angle measurement (discussed below) by the measurer in order to "direct", via memory flags, the activities of the RANG program in the "read" (INPUT) stage of that computation program. MANG tests, using AND's, bits 7,5, and 4 of Port B of the KIM for a high (5 volts) for buttons 3 (secondary done), 2 (primary done) and 1 (segment done) respectively, where reference is made to stages of measurement of the lepton angle in lepton induced inelastic, electromagnetic scattering interaction events in nuclear emulsion pellicle targets exposed to a high energy lepton beam. A typical event is seen in Figure 2. Vertical marks on the portion of the figure showing the incident primary lepton track separate x-coordinate "cells", which are typically chosen to be 50 microns in size for beam energies of around 150 GeV (giga electron volts) because the scattering angles are quite small (typically

TABLE 1. *System and Data Disk Files*

SECONDARY STORAGE:

Dual 5" Floppy Disks:

	File Number	Name	RAM Location	Track	Sector	File Number	Name	RAM Location	Track	Sector
	DIR 1		/DOS							
	01	%FM307	F300 FFFF	01	01	02	%TED	E000 EBCI	02	11
	03	%FMT	ED00 F0C9	04	03	04	%SOR	ED00 EE7C	04	11
	05	%BLD	ED00 F02B	04	14	06	%CPY	ED00 EDEA	05	05
	07	%DEL	ED00 ED89	05	07	08	%DIR	ED00 EDC6	05	09
	09	%*FM	ED00 EFEC	05	11	10	%REA	ED00 EDE2	06	01
	11	%PAK	ED00 EFD6	06	03	12	%LDN	ED00 EE48	06	09
	13	%NAM	ED00 EE13	06	12	14	%BLM	ED00 EF07	06	15
	15	%FRE	ED00 EE4F	07	04	16	%BASIC	2000 55FF	07	07
	17	%BAS	E000 EBFF	14	03	18	<BILIB	2000 38FF	15	11
Machine	19	%SNP	ED00 EE48	18	13	20	%VOICE	5E00 5FFF	18	16
Counter→	21	%MANG	9F00 9FFF	19	04	22	%BANG	9F00 9FFF	19	06
Data	23	@DANG	5000 6470	19	08	24	@CTEST	5000 51CC	22	01
Program	25	@DION	5000 5A4B	22	05	26	@SOUP	0000 0000	23	10
	27	@RTEST	4FF8 510c	23	13	28	@MUGET	5000 52A5	23	16
	29	@RANG	5000 6B80	24	06					
			\Angle Measurement Program					\Data Retrieval Program		
							Counter Test Program			
	DIR 2									
	01	@MUDAT	0000 0000	01	01					
			\Random Record Data File							

around ½ degrees). Thus, if we have completed one segment (after one "click" of the x-axis collar), and have pushed button 1, then the first BEQ branches to the section of MANG labeled PRIMARY DONE? within which section another BEQ branches to the section labeled SEGMENT, whence the AND #10 avoids LOOP (waiting point for no button pushed), and places, instead, a flag value of 01 to a safe place in RAM (here, $9FFA) into which the BASIC program can PEEK later on (to see if more segments should be read), whereupon, the 9 bytes of encoder data are "fetched" from the KIM data port (at $1700) and stored into RAM at $9FF0-$9FF8, and then return (RTS) is made to the FOR-NEXT loop in the BASIC program RANG. A close up of RANG's read in of the 9 bytes of segment data for a given point on the primary beam particle track is seen in Table 3, line 360. RANG goes to machine subroutine MANG, via BASIC'S jump to machine subroutine instruction USR(X), and proceeds to get 9 BCD bytes on each successive iteration (provided the "segment done" flag has been set so as to permit

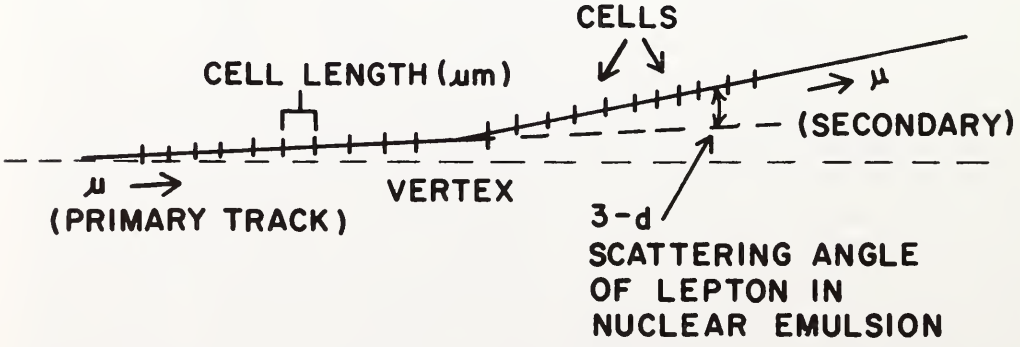

FIGURE 2. *Typical Lepton Track Scattering Topology*

TABLE 2. *Machine Subroutine % MANG*

```
                                    INITIALIZE:
9F00  D8              DECOFF        CLD              ; Clear Decimal
      A9 00           INIT          LDA #00          ; Clear Flags:
      8D FA 9F                      STA 9FFA         ; Segment Flag = 0
      8D FB 9F                      STA 9FFB         ; Primary Flag = 0
      8D FC 9F                      STA 9FFC         ; Secondary Flag = 0
      8D 01 17        MASK          STA 1701         ; Port A (all bits) = IN
      A9 OF                         LDY #0F          ; Port B bits 0-3 =OUT
                                                             bits 4-7 =IN
      8D 03 17                      STA 1703
      EA EA EA EA     DUMMY
                                    FLAGS:                    Flags Mask
                                                              IN  OUT
      AD 02 17        SECONDARY DONE? LDA 1702       ; Port B [76543210] 1702
                                                       Port A [76543210] 1700
                                                              IN
                                                             DATA
      29 80                         AND #80          ; Bit 7 (3rd Flag Button)
      F0 08                         BEQ Primary Done ; Go to Primary Done?
      A0 01                         LDY #01          ; Set Secondary Flag
      8C FC 9F                      STY 9FFC         ;
      4C 48 9F                      JMP FETCH        ; Get last ENCODER data
      AD 02 17        PRIMARY DONE? LDA 1702         ;
      29 20                         AND #20          ; Bit 5 (2nd flag button)
      F0 08                         BEQ Segment      ;
      A0 01                         LDY #01          ;
      8C FB 9F                      STY 9FFB         ; Set Primary Flag
      4C 48 9F                      JMP FETCH        ; Get last ENCODER data
9F36  AD 02 17        SEGMENT       LDA 1702         ;
      29 10                         AND #10          ; Bit 4 (1st flag button)
      F0 08                         BEQ LOOP         ; No button yet pushed
      A0 01                         LDY #01          ; Set Segment Flag
      8C FA 9F                      STY 9FFA         ;
      4C 48 9F                      JMP FETCH        ;
      4C 18 9F        LOOP          JMP SECONDARY    ; Wait for any flag button
                                        DONE?          to be pushed
                                    DATA FETCH:
      A2 00           FETCH         LDX #00          ; Mask to fetch first (X_LO) data
                                                       byte from counters
      8E 02 17        NEXT          STX 1702
      AD 00 17                      LDA 1700         ; Fetch data byte for track
                                                       segment
      9D F0 9F                      STA,X 9FF0
      E8                            INX              ; Increment byte fetch mask
      E0 09                         CPX #09          ; 9 byte yet?
      F0 02                         BEQ RETURN       ; Done fetching 9 bytes
      D0 F0                         BNE NEXT         ; Get next byte
      60              RETURN        RTS              ; Return to BASIC FOR-NEXT
                                                       Data Input Loop
```

a machine level fetch from the counters) of the FOR-NEXT loop until a PEEK at memory location 40955 ($9FFB) reveals a "primary done" flag value of 01 to have been set by the machine program (MANG) and detected by the PEEK in statement 400, at which time, transfer out of the FOR-NEXT loop is made and computations on the primary track portion of the event are begun. Thereafter, the same procedure is carried out for the secondary track portion of the event, at statement 510, with the loop escape made in 550 if the PEEK reveals that

TABLE 3. *Print Out of RANG Data Input Sections*

```
>LOAD"RANG", 1
>List  340,410
 340  INPUT"INPUT CELLP"; CP(K):CELL=CP(K):NN=0
 350  PRINT"INPUT YP,ZP PAIRS"
 360  FOR I=0 TO 100:X=USR(K):GOSUB 1790
 370  Y(I)=LD(3) +LD(4)*100+LD(5)*10000:REM PULSES
 380  Z (1) =(LD(6) + LD (7)*100 + LD(8)*10000)*100/UZ:REM MICRONS
 390  NN=NN+1
 400  IF PEEK (40955)=01 THEN 420
 410  NEXT I
>LIST 1790,1870
 1790 REM SUBROUTINE DEC TO HEX
 1800 PRINT
 1810 SPOT=40944
 1820 FOR J=0T08
 1830 LH=PEEK(SPOT+J)
 1840 LD(J)=INT(LH/16)*10+(LH-INT(LH/16)*16)
 1850 PRINT LD(J)
 1860 NEXT J
 1870 RETURN
>LIST 490,560
 490  INPUT"INPUT CELLS";CS(K):CELL=CS(K):NN=0
 500  PRINT"INPUT YS,ZS PAIRS"
 510  FOR I=0T0100:X=USR(K):GOSUB 1790
 520  Y(I)=LD(3)+LD(4)*100+LD(5)*10000:REM PULSES
 530  Z(I)=(LD(6)+LD(7)*100+LD(8)*10000)*100/UZ:REM MICRONS
 540  NN=NN+1
 550  IF PEEK(40956)=01 THEN 570
 560  NEXT 1
```

MANG has placed the "secondary done" value of 01 into memory location 40956 ($9FFC, see Table 2). The BASIC program RANG does not make use of the "segment done" flag to discontinue and exit the data INPUT loop like the "primary done" or "secondary done" flags do but rather uses it to alert RANG to continue with its INPUT loop iteration, i.e., RANG doesn't proceed to the next "FOR-NEXT" iteration if MANG doesn't first reach its "return from subroutine" (RTS) machine instruction unless the latter's SEGMENT section detects the first button's depression when an AND is made with bit 4 of the control port B of the KIM and a branch to FETCH is thus made in which the up/down digital counters are strobed for the 3 x-axis bytes, the 3 y-axis bytes, and the 3 z-axis bytes and the final return from subroutine (RTS) is thus made. Then the 9 BCD bytes, strobed to KIM port A and put by MANG into RAM at 9FF0-9FF8, are PEEK'd at by RANG's subroutine "DEC to HEX", statement 1830. Unfortunately, RANG's PEEK is programmed by BASIC's interpreter to convert HEX to DECIMAL, but the 9 counter bytes are already in BCD from the counters, so statement 1840 (see Table 3) reverses the PEEK'd values "back", i.e., "back" to BCD, whence the main program, at 370 and 380, assembles together the 3 y-axis bytes (Lo, Mi, and Hi) to a single decimal number N, i.e.,

$$N = (Lo)x(0)+(Mi)x(100)+(Hi)x(10{,}000) \text{ pulses.}$$

During an actual angle measurement, the BCD up/down counters are reset (see Figure 1) before starting either the primary or secondary for ease of data

printout checking. In any event, at any measurement point during an angle measurement, RANG checks to see if a count downwards from y = 0 pulses has been made and, if so, RANG resets 0 to 1,000,000 pulses so that ex., an incorrect "first difference" in the y-axis coordinates of 0-999,900 = -999,900 pulses is replaced by the correct difference of 999,900-1,000,000 = -100 pulses. The eyepiece microfilometer cross hair scale, seen in the eyepiece view, visually denotes the y value, in mm, of the movable horizontal member of the eyepiece cross hair (1) as the operator has translated the x-axis screw of the KORSTKA microscope stage over one x-segment (one "cell length" or "click" of the notched x axis sleeve) of the primary or secondary track of the sub-nuclear interaction, adjusting the z-axis control to keep the track portion, being moved along, in focus.

Since one turn of the z-axis microscope collar amounts to 100 microns but one turn of the attached optical encoder amounts to precisely 2000 pulses, then RANG converts (see line 380 of Table 3 for the case of the primary track, and 530 for the case of the secondary track) the z-measurement to

$$z(\text{microns}) = z(\text{pulses})*100 \text{ microns/rev/UZ}$$

where UZ = 2000 pulses/rev. The y-axis measurements are based on a 1440 pulse/rev encoder and the x-axis on a 2000 pulse/rev encoder. The x-axis information is not used for the angle measurement algorithm since the x-cell length advance is changed as frequently as twice per event (once for the primary and possibly again for the secondary) and can be easily done manually on the x-axis collar. Its numerical value forms part of the initially typed in demographic data for this particular angle algorithm.

Table 4 shows the Memory Map chosen for the KIM 6502 based system. We see that the 12K HDE microsoft BASIC workspace begins at $5000, leaving 20K of RAM for the computation program RANG's workspace and its machine

TABLE 4. *KIM System Memory Allotment Map*

PRIMARY STORAGE:		
Memory Board:	Hex Address Range:	Content of Function:
8K RAM	FFFF E000	Disk Operating System
	C000	
16K EPROM	B000 A000	Disk Boot, Extended Monitor
	9000 Machine:	
	8000	
16K RAM	7000	Work Space: Machine & Basic
	6000 Basic:	
	5000	
	4000	
16K RAM	3000 2000	Disk BASIC: HDE Microsoft
8K RAM & ROM (CPU Board)	1000 0000	Disk Port CPU Monitor, Stack User RAM, Ports, Timers

subroutine MANG which was placed in RAM starting at $9000 with its 9 bytes of data and 3 flag bytes starting at $9F00. The Disk Operating System by HDE facilitates secondary (disk) storage of each event's principal results along with its deomgraphic data into the random record data file labeled MUDAT (see Table 1). For ease of the event's data retrieval, the record number that is to hold the principal event data is set by RANG to the event number of the scattering event being measured. Thus, data may be retrieved for several previously measured events in a systematic way by some other programs written in BASIC such as are needed, for example, to compare experiment with theory predictions, etc.

A typical session begins by warming up the microscope booting up the KIM system, loading BASIC, running the interactive program RANG up to the first data prompt, centering the eyepiece crosshair, locating and bringing into the center of the eyepiece's field of view the "star" whose beam particle track's scattering angle is to be measured, accessing the event demographics and the topography of the scattering beam track (primary and secondary sections), typing into the TTY the demographics in response to RANG's first prompt, and finally, positioning a point several cells (in x) upstream along the incident primary beam track section from the vertex into the center of the eyepiece field of view by moving the x, and y (eyepiece microfilometer x-axis cross hair) and z controls until the cross hair lateral axis lies coincident with one field of view's worth of track grains (the cross hairs are rotatable about an axis optically concentric to the z-direction such that the lateral, internally movable cross hair can be very well lined up with the general direction of the usually slanted event primary or secondary track). Then, the reset button is pushed followed by a push of the first button ("segment done") whence a series of nine zero bytes is printed by RANG to the TTY for these initial x,y,z coordinates. Then, while the microscope stage's x-coordinate is advanced one x-cell length (usually the 50 micron knotch is used at 150 billion electron volt beam energies) closer to the vertex, the z (focus) and y (crosshair)is readjusted until the downstream end of the first cell length (or segment) is reached at which time button 1 ("segment done" button) is pushed whereupon RANG outputs the advance in y and z in terms of raw coordinate data (BCD pulse counts). When the last of the primary's cell lengths is finished (the upstream side of the vertex will now be in view; see left side of Figure 2), button 2 ("primary done") is depressed signaling RANG to make calculations of the primary track angle elements and print them to the TTY. Then RANG has the TTY prompt the measurer with the bell to type in the cell length of the secondary section (scattered section) of the scattering particle track (usually the same value as the primary cell length).

Then the stage is moved to a new initial position just downstream from the vertex and in view of the vertex, whence the reset button is pushed and then the first ("segment done") button is pushed getting a set of nine zero bytes on the TTY. The cross hair is set along the secondary at several positions downstream (each separated by the chosen x-cell length) and pushes of the first button ("segment done") are made as with the primary. When the last position is reached, the third button ("secondary done") is depressed whence RANG now calculates and prints the secondary's angle elements and then goes on to find the net 3-d scattering "space angle" (as well as the next x-y projection angle and the net r-z "dip angle" where r is the projected horizontal displacement of a segment). Then RANG has the TTY print a prompt asking if a total print out of the detailed measurement results is desired. At this point, the measurer may feel that he has not exercised quite enough care in measuring the coordinates or a quick perusal of the print out might tell him this and so when he is asked next if he

wishes to retain this measurement, he will type no in response, whence RANG's running average of angle results will not include that particular measurement. Then, when he is asked next if he wishes to re-measure, he might do so if he wishes to increase his precision statistics on that event further, or he may not do so. When he has done his last re-measurement, RANG computes the grand average of that event's principle angle results and demographic data and stores them to the record in the random storage data disk (in the applications drive) file MUDAT (see TABLE 1) that has the same number as the event itself. At this point, one can close the session (RANG closes out the data file MUDAT) or begin a new event.

Results and Conclusions

The system works quite well. Before, it was not time feasible to make several re-measurements (whether to correct marginal measuring techniques or to increase precision statistics) on a given event, because hundreds of such events might need to be measured before comparison with theory predictions can be made meaningful. But now, the measurement time has been reduced from upwards to an hour (for just one measurement) down to 15 minutes (including several re-measurements if wished). Further, the automation in data taking has greatly enhanced the reduction of error in contrast to before when there were reading errors (parallax, etc.), copying errors, typing errors, etc., not to mention the time required for all these latter tasks.

It is hoped that future use of this system for other types of measurement algorithms in the context of the nuclear emulsion technique of elementary particle research, whether using y and/or z and/or x dimension optical encoders, will only require a modification of the calculational source program (like RANG here), not the hardware.

Literature Cited

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PLANT TAXONOMY

Chairman: DONALD R. HENDRICKS
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801 Elks Road, Richmond, Indiana 47374

Chairman-Elect: JAMES R. ALDRICH
Indiana Department of Natural Resources
612 State Office Building, Indianapolis, Indiana 46204

ABSTRACTS

Hybridization between Elymus virginicus and E. hystrix. JOHN E. EBINGER, Department of Botany, Eastern Illinois University, Charleston, Illinois 61920.——*Elymus virginicus* and *E. hystrix* are very distinct, wide ranging species that are common in upland wooded habitats throughout eastern North America. An analysis of specimens from areas where both species occur indicate that they commonly hybridize. The results of this analysis plotted on scatter diagrams show that F_1 hybrids commonly occur in disturbed habitats, and that these hybrids occasionally backcross with the parent species. These hybrids, which are usually sterile, are intermediate in their characteristics showing considerable variation in glume structure and partially spreading spikelets.

Numerical Classifications: Cladistics or Phenetics? RICHARD J. JENSEN, Department of Biology, Saint Mary's College, Notre Dame, Indiana 46556.——The construction of a classificatory system by numerical taxonomic methodologies requires an *a priori* conception of what it is that the classification should represent. Phenetic classifications, based on overall similarity, have been argued to best represent natural groups. These classifications are not intended to present precise views of phylogeny, but may be used as close approximations. Cladistic classifications, based on the recognition of monophyletic groups, are intended to accurately represent phylogenetic history. Cladists argue that their classifications are, therefore, more natural than phenetic classifications. Pheneticists and cladists perform similar operations in the initial stages of constructing a classification. Cladistics, however, requires the estimation of character state polarities and it is argued here that this introduces a significant source of error not found in phenetic classifications. The conclusion is that general classifications should be based on phenetic criteria and that cladistic methods be employed to construct hypotheses of evolutionary relationships, but not as a means of constructing classifications. An example, based on analyses of American oaks, is presented to demonstrate the complementary use of both methods.

A Unique Ruderal Plant Association in the Central Panhandle of Florida. GAYTON C. MARKS, Department of Biology, Valparaiso University, Valparaiso, Indiana 46383.—— A most unlikely plant association has become established in a Walton County highway ditch near Freeport, Florida.

These plants probably should be considered opportunists as they have taken advantage of wet soil, full sun, an unlimited supply of insects and ideal growing temperatures.

Nine species of carnivorous plants can be identified. Eight of these are insectivorous. Four genera of three families are found here in great numbers. These

include two species of pitcher plant, *Sarracenia*; three species of sundew, *Drosera*; three species of butterworts, *Pinguicula*; and but a single species of bladderwort, *Utricularia subulata*.

Their beauty in flower is enhanced by several colorful orchids, gaudy milkworts, composites, sedges, grasses, many aquatic or semiaquatic monocots and a single species of a clubmoss, *Lycopodium*. Anthesis is primarily limited to spring but a few species tardily exhibit flowering into summer and autumn.

The singularity of such a plant community is truly a delight for the northern botanist.

Glades and Barrens of Crawford and Perry Counties, Indiana

John A. Bacone
and
Lee A. Casebere
Division of Nature Preserves
Indiana Department of Natural Resources
Indianapolis, Indiana 46204
and
Max D. Hutchison
Belknap, Illinois 62908

Introduction

Glades and barrens are natural openings within the forest in southern Indiana. The openings termed glades are those with bedrock at or near the surface, and are usually on steep south, southwest, or west-facing slopes. In southern Indiana, remnant glades occur most commonly on limestone (2), and this is the case in Crawford and Perry Counties. However, sandstone glades are known to occur in other states in the Midwest (26), and one remnant was found in Perry County.

Barrens are found on relatively deep soils, sometimes with very little slope. In presettlement times, they were rather extensive in portions of southern Indiana (16), but apparently quickly disappeared following settlement (9). This natural community has not been previously described in studies of Indiana's extant plant communities (15,20).

In this paper, barrens, limestone glades, and the sandstone glade, all of which were recently located in Crawford and Perry Counties, will be discussed. The methods used to locate and evaluate them are also discussed, and preliminary species lists for each community are presented.

Methods

A systematic search for glade and barren remnants was made in Crawford and Perry Counties during 1981 and 1982 (13,14). The methods used were similar to those used to find glades in Harrison County, Indiana (1,12). The most recent aerial photographs available at the Agriculture Stabilization and Conservation Service offices in each county were examined, in conjunction with the 7.5 minute United States Geological Survey topographic quadrangle maps. Openings in the forest were scrutinized in an effort to locate those with potential to be natural. Openings that were obviously man-made were eliminated from consideration. Older aerial photographs were examined to further evaluate the past history of these openings.

Each potential natural opening was then checked from the air. A high-wing Cessna aircraft was used, at an altitude of 1,000 feet. The flight took place following the growing season, so that vegetation could be analyzed using the characteristic coloration of the native prairie grasses as compared to old world flora. The sites with high potential were then examined on the ground.

During the ground survey, a number of significant sites were found. These included one sandstone glade and a number of both barrens and glades. These areas were determined to be of high natural quality based on such factors as the relative absence of alien species, a high diversity of native species, a lack of woody encroachment, and the lack of obvious signs of disturbance.

Results

One sandstone glade was located in southern Perry County. This community occupied a long, narrow strip at the top of a 150 foot high cliff. Vegetation is sparse, as bedrock covers much of the area. The limited amount of soil is of the Muskingum-Gilpin Association (21). Lichens, mosses, and clumps of grasses and forbs are scattered throughout. Gnarled cedars (*Juniperus virginiana crebra*) and oaks, including post oak (*Quercus stellata*) and black jack oak (*Q. marilandica*) attest to the xeric environment. Thirty-two native taxa were identified. (Table 1). Broomsedge (*Andropogon virginicus*), little bluestem (*A. scoparius*), and *Danthonia spicata* are the most common grasses. *Tephosia virginiana* and *Viola pedata* are the most common forbs. Shrubs are also common here, including mountain laurel (*Kalmia latifolia*), and three species of *Vaccinium* (*arboreum, stamineum,* and *vacillans*).

A limestone glade was also located in Perry County. In contrast to Harrison County glades (1) it has deeper soils. It is on south-southwest facing slopes, with limestone outcropping. Soils here are Gilpin, Wellston and Muskingum (21). Indian grass (*Sorghastrum nutans*), big bluestem (*Andropogon gerardi*) and little bluestem are common grasses. A rich diversity of forbs, including *Eryngium yuccifolium*, *Helianthus mollis*, *Gentiana quinquefolia* and *Liatris aspera* also occur here. Forty-seven native taxa were found here (Table 1).

The barrens and glades in Crawford County were found on silty clay loam and silt loam soils (Hagerstown, Wellston and Zanesville) (25). The glades had deeper soils than those in Harrison County (1). Some erosion was evident. Species composition was similar to that of the Perry County glade. However, over 100 native taxa were found, probably due to the higher number of sites located. On the barrens, prairie dock (*Silphium terebinthinaceum*) was very common, and Elliott's bluestem (*Andropogon elliottii*) was an important grass, in addition to big bluestem, little bluestem, and Indian grass.

A number of species considered rare in Indiana (3,4) were found in these communities. In addition, ten of the taxa reported from Perry County sites and 25 from Crawford County sites were not reported by Deam (7). Thrity-nine of the taxa were also reported to occur on the limestone glades located in nearby Harrison County. This information is presented in Table 1.

Discussion

The glades and barrens presented in this paper are presumably remnants of the presettlement barrens that covered much of southern Indiana (7,9,16). The presettlement barrens of Harrison and Washington Counties were primarily restricted to the Mitchell Plain Physiographic Province (16), where a number of remnant limestone glades were recently described (1). No remnant barrens were located in those counties. The barrens and limestone glades described in this paper are found west of the Mitchell Plain, in the Crawford Upland Province (18), indicating presettlement barrens may have been widespread.

Except for historical accounts, barrens and glades have been previously undescribed and unmapped in southern Indiana (11,15,17,20,23) until recently (1,16). In the early accounts (6), the term "barrens" probably included both the glade and the barren communities. Charles Deam was aware that presettlement barrens once existed in Indiana (9) but apparently never located any (7). This is further evidenced by the fact that several praire species, including *Eryngium yuccifolium*, *Helianthus mollis*, *Silphium terebinthinaceum* and *Solidago rigida* were not reported from Crawford County in his 1940 *Flora of Indiana* (7).

TABLE 1. *A preliminary species list for glades and barrens of Crawford and Perry Counties, Indiana.*[4]
* = *on the "Preliminary list of endangered and threatened vascular plants of Indiana"* (3,4)

Taxon	Sandstone Glade Perry County	Limestone Glade Perry County	Glade/Barren Crawford Co.
Acer rubrum [1]	x		x
Agave virginica [3]		x	x
Amelanchier sp.	x		
Andropogon elliottii [2]			x
Andropogon gerardii [2,3]		x	x
Andropogon scoparius [3]	x	x	x
Andropogon virginicus	x		x
Anemone virginiana [3]			x
Angelica venenosa			x
Antennaria plantaginifolia	x		
Apocynum cannabinum [2]		x	x
Aristida longespica [1]	x		
Aronia melanocarpa			x
Asclepias tuberosa			x
Asclepias verticillata [3]		x	x
Asclepias viridiflora [2]			x
Ascyrum hypericoides			x
Aster laevis [2]		x	x
Aster novae-angliae [2]			x
Aster patens [3]			x
Aster puniceus [2]			
**Aster solidagineus*			x
Blephilia ciliata		x	
Carya ovata			x
Cassia fasciculata		x	x
Ceanothus americanus			x
Celtis pumila			x
Cercis canadensis [3]		x	x
Convolvulus spithamaeus			x
Coreopsis tripteris [3]			x
Cornus florida		x	x
Corylus americana			x
Crataegus sp.			x
Cunila origanoides	x		
Danthonia spicata [3]	x		x
Desmodium rotundifolius			x
Diodia teres setifolia		x	
Diospyros virginiana	x		x
Eleocharis compressa			x
Eryngium yuccaefolium [1,2,3]		x	x
Eupatorium fistulosum		x	
Euphorbia corollata [3]	x	x	x
Fagus grandifolia	x		x
Fraxinus americana			x
**Galactia volubilis mississippiensis* [2,3]			x
Gentiana quinquefolia [1,3]		x	
Gerardia tenuifolia [3]			x
Helianthus divaricatus		x	x
Helianthus hirsutus			x
Helianthus mollis [1,2]		x	x
Helianthus occidentalis			x

1. Taxon not reported for Perry County in *Flora of Indiana* (7).
2. Taxon not reported for Crawford County in *Flora of Indiana* (7).
3. Taxon also reported from limestone glades in Harrison County, Indiana (2).
4. Nomenclature according to Gleason and Cronquist (10).

TABLE 1.—Continued

Taxon	Sandstone Glade Perry County	Limestone Glade Perry County	Glade/Barren Crawford Co.
**Hypericum dolabriforme*			x
Hypericum gentianoides	x		
Hypericum prolificum			x
Hypericum punctatum			x
Juniperus virginiana crebra [3]	x	x	x
**Kalmia latifolia*	x		
Krigia dandelion	x		
Lechea tenuifolia	x		
Lespedeza capitata [1,3]		x	
Lespedeza cuneata			x
Lespedeza hirta			x
Lespedeza procumbens		x	x
Lespedeza virginica			x
Liatris aspera [3]		x	x
Liatris spicata [3]			x
**Liatris squarrosa* [1,3]	x	x	x
Linum medium [1]	x	x	x
**Linum sulcatum* [3]			x
Linum virginicum			x
Lithospermum canescens [3]		x	x
Lobelia spicata [2,3]		x	x
Lysimachia quadrifolia			x
Monarda clinipodia			x
Monarda fistulosa [2,3]		x	x
Ostrya virginiana [3]		x	x
Panicum depauperatum [1]	x		
Panicum sp.			x
Phlox pilosa [2,3]		x	x
Physostegia virginiana [2,3]		x	x
Polygala verticillata [2]			x
Potentilla simplex argyrisma	x	x	x
Psoralea psoralioides eglandulosa			x
Pycnanthemum flexuosum		x	x
Pycnanthemum pycnanthemoides			x
Quercus alba	x	x	x
Quercus coccinea [1]	x		x
Quercus imbricaria			x
Quercus marilandica	x	x	x
Quercus muhlenbergii [3]			x
Quercus prinus	x		
Quercus velutina			x
Ratibida pinnata [2,3]		x	x
Rhamnus caroliniana [3]		x	x
Rhus aromatica			x
Rhus copallina			x
Rhus glabra	x		x
Robinia pseudo-acacia	x		
Rosa carolina [3]		x	x
Rudbeckia fulgida [2,3]			x
Rudbeckia hirta		x	x
Ruellia caroliniensis [3]		x	x
Sabatia angularis [3]		x	x
Sassafras albidum	x	x	x
Scirpus lineatus			x
Scleria sp.			x
Senecio obovatus [2]			x

1. Taxon not reported for Perry County in *Flora of Indiana* (7).
2. Taxon not reported for Crawford County in *Flora of Indiana* (7).
3. Taxon also reported from limestone glades in Harrison County, Indiana (2).

TABLE 1.—Continued

Taxon	Sandstone Glade Perry County	Limestone Glade Perry County	Glade/Barren Crawford Co.
Silphium integrifolium [1]		x	
Silphium terebinthinaceum [2]		x	x
Silphium trifoliatum [3]		x	x
Sisyrinchium albidum [3]			x
**Smilax bona-nox* [3]			x
Smilax sp.			x
Solidago graminifolia			x
Solidago juncea [2]			x
Solidago rigida [1,2]		x	x
Sorghastrum nutans [3]		x	x
Sporobolus vaginiflorus [3]			x
Strophostyles umbellata			x
Stylosanthes biflora [2]			x
Swertia carolinensis [3]		x	
Tephrosia virginiana	x		x
Thaspium trifoliatum [2]			x
Ulmus alata			x
**Vaccinium arboreum*	x		
Vaccinium stamineum	x		x
Vaccinium vacillans	x		
Viburnum rufidulum [3]		x	
Viola pedata	x		
**Zizea aptera* [2]			x
Totals	32	47	108

1. Taxon not reported for Perry County in *Flora of Indiana* (7).
2. Taxon not reported for Crawford County in *Flora of Indiana* (7).
3. Taxon also reported from limestone glades in Harrison County, Indiana (2).

Apparently, the barrens as a major plant community in Indiana disappeared quickly following settlement. The disappearance of this community is attributed primarily to cultivation and fire suppression (6,9,16,19).

The limestone glades and barrens remnants described in this paper are rich and prairie-like in both species composition and appearance. They are not very similar to those described for Illinois by Vestal (24), who spoke of "a high proportion of forest herbs, and lacking many prairie plants." Rather they resemble those described in Kentucky (8), as a "beautiful meadow, where grass was from two to three feet high" (19). Collett (6) described the barrens of south-central Indiana as "a nearly typical prairie exhibiting a few gnarled and scotched shrubs, and covered with a luxuriant growth of tall prairie grasses, herbs and vines." They have a high affinity to the prairies and savannas of northwest Indiana—sixty taxa recorded here are found in those communities (22).

The remnants described here appear to be of a high quality, based on the diversity and lack of aliens. It is probably impossible to ascertain why they still remain today, but possible explanations may include all or a combination of the following: mowing, grazing, erosion, sterility of soils (2), or succession from agriculture undertaken very long ago (5).

Floristically, the sandstone glade is quite different from the other glades and barrens. The differences include a lack of Indian grass, very few forbs and the presence of a variety of ericaceous shrubs.

The glades and barrens really cannot be separated floristically. The main difference was visual, with deeper soils and the presence of prairie dock

distinguishing the barrens. These communities, although seperated by geographic distance and man-made barriers today, were probably very interrelated during presettlement times.

Protection and management of these communities will be necessary if they are to continue to exist in Indiana. Management needs include both prescribed burning, and cutting of woody speices. The aerial photographs show a remarkable decrease in size in the last forty years due to encroachment by woody species. Erosion is also a threat in some of these areas. Several of the barren remnants have recently been dedicated as an Indiana State Nature Preserve, which will provide legal protection. Other glade and barren remnants are in public ownership within Harrison-Crawford State Forest and the Hoosier National Forest. Hopefully, these and some of those in private ownership will be also given protection.

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Vascular Plants of Bendix Woods Nature Preserve St. Joseph County, Indiana

THOMAS D. BLODGETT, Bendix Woods County Park
New Carlisle, Indiana 46552
and
VICTOR L. RIEMENSCHNEIDER, Department of Biological Sciences
Indiana University at South Bend, South Bend, Indiana 46634

A floristic inventory of the Bendix Woods Nature Preserve was undertaken by the authors as part of a larger study (3). The study began in March 1981. The natural area, 4.8 km south of New Carlisle, Indiana, is located in the east one-half of the south portion of Bendix Woods County Park, Section 11, Township 37 North, Range 1 West, Lydic Quadrangle. The Preserve and surrounding area are part of the kame facies of the Largo Formation of the Wisconsinan Glacial period. The soils are of the Hillsdale type, well drained, sandy loam, level to strongly sloping (5). The original vegetation was mixed hardwoods.

A 12.2 ha natural area in the Park was identified by Lindsey et al. (2) as a unique, old growth, hardwood forest. Subsequently, 10.9 ha was dedicated as a State Nature Preserve in 1971, owned and administrated by the St. Joseph County Parks and Recreation Department. The Preserve and peripheral environs has been protected since 1926.

Methods

During the growing seasons of 1981 and 1982 the authors divided the Preserve into 50 m X 50 m, quarter hectare quadrats. All woody species greater than 4 cm dbh were recorded for each quadrat (3). Sedges, grasses, ferns and herbs in flower were also recorded.

Additionally, 100 random circular (r = 60 cm) herbaceous study plots were established. The plots were surveyed either in the spring or fall, or in both seasons of 1981 and 1982. All taxa less than 1 meter in height were recorded.

Species were also noted and, if in flower, collected whenever the authors hiked the Preserve's trails. Information recorded included canopy condition (open or closed), associated species, and location relative to a corner of a quadrat or herbaceous plot.

Results and Discussion

To date, 119 species, representing 90 genera, have been identified in the Preserve. Of this total 6 taxa are ferns and fern allies, 11 grasses and sedges, 59 species of flowering herbs, 14 shrub and woody vine taxa, and 29 tree species. Native taxa number 114; 5 are introduced (4).

To properly evaluate the quality of natural areas within the 22 county Chicago Region, Swink and Wilhelm (4) have developed an Evaluation Checklist and a Rating Index. Taxa found in the Region have been assigned numerical values—ratings—based upon the taxon's frequency and faithfulness to various synecological parameters. The 0 rating implies that a species grows under a variety of conditions; they are omnipresent. A 5 rating means the taxon is less common and typical of more advanced seral stages. A 10 rating is assigned to those taxa associated with near-climax conditions and a restricted set of synecological parameters. Taxa that are quite rare or endangered in the Region are assigned values of 15 and 20 respectively.

To determine an area's natural quality, a Rating Index is calculated. The Rating Index is determined mathematically using the following formula: $I = R/\sqrt{N}$, where I = Rating Index, R = the sum of the numerical ratings for all native taxa recorded for the area, and N = the number of recorded taxa (4). In the Chicago Region, Swink and Wilhelm (4) considered areas with an Index of 50 or higher as "extremely rare". The Index for the Bendix Woods Nature Preserve is 69.45, using R = 739 and N = 113 (native species).

According to the Evaluation Checklist of Swink and Wilhelm (4), seventeen species found in Bendix Woods are rated as endangered or rare. The endangered species are (follows the nomenclature of Swink and Wilhelm) *Aplectrum hymale*, *Dentaria diphylla* and *Hydrophyllum canadense*. In Bendix Woods, *D. diphylla* is thriving in several locations and *H. canadense* is abundant.

The 14 taxa classified as being rare are: *Asimina triloba*, *Athyrium pycnocarpon*, *A. thelypterioides*, *Carex careyana*, *C. hitchcockiana*, *C. plantaginea*, *Dryopteris goldiana*, *Monotropa uniflora*, *Morus rubra*, *Orchis spectabilis*, *Panax quinquefolis*, *Sanicula trifoliata*, *Stylophorum diphyllum* and *Viola canadensis*.

Rated as 10—taxon indicative of an advanced seral to climax stage and of a narrow range of ecological conditions—are 9 taxa. Included in this group are *Carex albursina*, *Fagus grandifolia*, *Geranium robertianum* and *Sambucus pubens*.

Carpinus caroliniana, *Cornus alternifolia*, *Floerkea proserpinacoides*, *Gymnocladus dioicia*, *Hydrophyllum apendiculatum*, and *Juglans cinera* are some of the 19 species rated as 8 or 9. Another 38 taxa are rated as 5, 6 or 7. A complete list of species is presented in Table 1. However, we are continuing our studies of Bendix Woods Nature Preserve. Individuals may write the authors for a current list.

Two taxa are on the "Preliminary List of Endangered and Threatened Vascular Plants in Indiana" (1): *Geranium robertianum*, endangered, and *Dentaria diphylla*, threatened.

Actaea pachypoda, *Athyrium pycnocarpon*, *Dentaria diphylla*, and *Dryopteris goldiana* are new species records for St Joseph County. Voucher specimens have been collected and will be deposited in the Indiana University Deam Herbarium.

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TABLE 1. *Vascular Plants of Bendix Woods Nature Preserve*

1. *Acer nigrum* Michx.f.
2. *Acer saccharum* Marsh.
3. *Actaea pachypoda* Ell.
4. *Ailanthus altissima* (Mill) Swingle
5. *Allium tricoccum* Ait.
6. *Amphicarpa bracteata* (L.) Fern.
7. *Aplectrum hyemale* (Muhl.) Torr.
8. *Arisaema atrorubens* (Ait.) Blume.
9. *Asarum canadense* L.
10. *Asimina triloba* (L.) Dunal.
11. *Aster lowrieanus* Porter
12. *Athyrium pycnocarpon* (Spreng.) Tidestr.
13. *Athyrium thelypterioides* (Michx.) Desv.
14. *Berberis thunbergi* DC.
15. *Botrychium virginianum* (L.) Sw.
16. *Campanula americana* L.
17. *Carex albursina* Sheldon.
18. *Carex careyana* Torr.
19. *Carex hirtifolia* Mackenz.
20. *Carex hitchcockiana* Dew.
21. *Carex jamesii* Lam.
22. *Carex laxiflora* Lam.
23. *Carex plantaginea* Lam.
24. *Carex sparganoides* Muhl.
25. *Carpinus caroliniana* Willd.
26. *Carya cordiformis* (Wang.) K. Koch.
27. *Caulophyllum thalictroides* (L.) Michx.
28. *Celtis occidentalis* L.
29. *Circaea quadrisulcata* (Maxim.) French & Sav.
30. *Claytonia virginica* L.
31. *Cornus alternifolia* L.f.
32. *Crataegus* sp.
33. *Cryptotaenia canadensis* (L.) DC.
34. *Cystopteris fragilis* (L.) Bernh.
35. *Dentaria diphylla* Michx.
36. *Dentaria laciniata* Muhl.
37. *Desmodium glutinosum* (Muhl.) Wood
38. *Dicentra canadensis* (Goldie) Walp.
39. *Dicentra cucullaria* (L.) Bernh.
40. *Dryopteris goldiana* (Hook.) Gray
41. *Dryopteris spinulosa* (O.F.Muell) Watt.
42. *Epifagus virginiana* (L.) Bart.
43. *Erigenia bulbosa* (Michx.) Nutt.
44. *Erythronium americanum* Ker.
45. *Euonymus obovatus* Nutt.
46. *Fagus grandifolia* Ehrh.
47. *Festuca obtusa* Biehler
48. *Floerkea proserpinacoides* Willd.
49. *Fraxinus americana* L.
50. *Fraxinus pennsylvanica* Marsh
51. *Fraxinus quadrangulata* Michx.
52. *Galium aparine* L.
53. *Galium triflorum* Michx.
54. *Geranium maculatum* L.
55. *Geranium robertianum* L.
56. *Geum canadense* Jacq.
57. *Gymnocladus dioica* (L.) K. Koch
58. *Hepatica acutiloba* DC.
59. *Hydrophyllum appendiculatum* Michx.
60. *Hydrophyllum canadense* L.
61. *Hydrophyllum virginianum* L.
62. *Impatiens pallida* Nutt.
63. *Isopyrum biternatum* (Raf.) T&G.

TABLE 1.—Continued

64. *Juglans cinerea* L.
65. *Juglans nigra* L.
66. *Lindera benzoin* (L.) Blume.
67. *Liriodendron tulipifera* L.
68. *Monotropa uniflora* L.
69. *Morus rubra* L.
70. *Orchis spectabilis* L.
71. *Osmorhiza claytoni* (Michx.) C.B. Clarke
72. *Osmorhiza longistylis* (Torr.) DC.
73. *Ostrya virginiana* (Mill.) K. Koch
74. *Oxalis stricta* L.
75. *Panax quinquefolis* L.
76. *Parthenocissus quinquefolia* (L.) Planch
77. *Phlox divaricata* L.
78. *Phryma leptostachya* L.
79. *Phytolacca americana* L.
80. *Pilea pumila* (L.) Gray
81. *Platanus occidentalis* L.
82. *Poa sylvestris* Gray
83. *Podophyllum peltatum* L.
84. *Polygonatum pubescens* (Willd.) Pursh.
85. *Populus deltoides* Marsh.
86. *Prunus serotina* Ehrh.
87. *Prunus virginiana* L.
88. *Pyrus malus* L.
89. *Quercus rubra* L.
90. *Ranunculus abortivus* L.
91. *Ranunculus recurvatus* Poir.
92. *Rhus radicans* L.
93. *Ribes cynosbati* L.
94. *Rosa multiflora* Thunb.
95. *Rubus allegheniensis* Porter
96. *Rubus occidentalis* L.
97. *Sambucus canadensis* L.
98. *Sambucus pubens* Michx.
99. *Sanguinaria canadensis* L.
100. *Sanicula trifoliata* Bickn.
101. *Sassafras albidum* (Nutt.) Nees
102. *Scrophularia marilandica* L.
103. *Setaria viridis* (L.) Beauv.
104. *Smilacina racemosa* (L.) Desf.
105. *Smilax taminoides* var. *hispida* (Muhl.) Fern.
106. *Solanum dulcamara* L.
107. *Staphylea trifolia* L.
108. *Stylophorum diphyllum* (Michx.) Nutt.
109. *Tilia americana* L.
110. *Tovaria virginiana* (L.) Raf.
111. *Trillium flexipes* Raf.
112. *Trillium grandiflorum* (Michx.) Salisb.
113. *Trillium recurvatum* Beck
114. *Ulmus americana* L.
115. *Ulmus rubra* Muhl.
116. *Urtica dioica* L.
117. *Viola canadensis* L.
118. *Viola pensylvanica* Michx. (f).
119. *Viola sororia* Willd.

Additions to the Flora of Southern Indiana

MICHAEL A. HOMOYA
Indiana Natural Heritage Program, Indiana Department of Natural Resources
Indianapolis, Indiana 46204

Introduction

Considerable new information has been added to the knowledge of southern Indiana's flora due to field activities by members of the Indiana Natural Heritage Program. During 1982 four species new to Indiana were recorded, as well as two species new to southern Indiana. Thirteen infrequently observed species were also noted at new locations.

All collection numbers are those of the Indiana Natural Heritage Program. Voucher specimens are to be deposited in the Deam Herbarium of Indiana University at Bloomington. The nomenclature follows Fernald (5) with the exception of *Platanthera*, with follows Luer (7) and *Lorinseria* and *Lycopodium*, which follows Cranfill (2).

Species New to Indiana

Itea virginica L. That this shrub was unknown from Indiana prior to our discovery is surprising, especially considering that we have collected it from three different sites. It may have been overlooked by early botanists (a good possibility, for all three sites are in somewhat remote areas), or there may have been a recent range extension into the state from the south. It occurs in a seep spring near Haysville (May 24 *Homoya, Casebere, and Hedge 4034*) and in a lowland forest at Buffalo Flat near Jasper (November 9 *Homoya and Post s.n.*), both in Dubois County. We also have it from a sinkhole swamp near Central Barren in Harrison County (July 14 *Homoya and Hedge 4216*). Associates include *Liquidambar styraciflua*, *Quercus michauxii*, *Nyssa sylvatica*, *Bignonia capreolata*, *Carex gigantea*, and *Peltandra virginica*.

Lorinseria areolata (L.) Presl. After observing a major range extension of this fern into southern Illinois during the past fifteen years, its discovery in southern Indiana was not totally unexpected. On June 24 we found two small populations growing on the edge of a sinkhole swamp southeast of Palmyra in Harrison County *(Homoya, Aldrich, Post, and Starcs 4072)*. The most frequent habitats used by *Lorinseria* in southern Illinois are sandstone cliffs; it will be interesting to see if similar environments are eventually utilized in Indiana.

Panicum scoparium Lam. This large grass is readily identified by its plush, velvety pubescence of culms and leaves and its glabrous, viscid band below each bearded node. A southern species of damp thickets and fields, it was first discovered in nearby southern Illinois in 1967 (8) and is now known from Posey County, Indiana. It has been reported for Indiana by several collectors, but was excluded by Deam due to error in identification (3,4). Several large colonies were found growing in an open, seasonably wet "flat" approximately two miles west of Hovey Lake Fish and Wildlife Area (August 9 *Homoya and Aldrich s.n.*).

Trichomanes boschianum Sturm. This temperate member of the mostly tropical *Hymenophyllaceae* was found growing in a small, dark grotto at the base of a sandstone cliff near Yellow Birch Ravine Nature Preserve in Crawford County (May 22, 1977 *Mark and Julius Swayne s.n.*). An intensive search in similar habitats in nearby areas has failed to reveal new sites for the filmy fern, making it one

of the rarest plants in Indiana. In addition, the severe winters subsequent to its discovery by the Swaynes have reduced the existing population considerably to only twenty to thirty small fronds *(fide* Homoya, J. and S. Aldrich, Post, C. and M. Ritter from observation on February 27, 1982).

Species New to Southern Indiana

Scleria pauciflora Muhl. ex Willd. Previously known only from northwestern Indiana, we now have two sites for it from the extreme southern part of the state. Several plants were found growing in a xeric upland forest near Hindostan Falls in Martin County with *Quercus prinus. Q. marilandica, Vaccinium arboreum, Danthonia spicata,* and *Lechea tenuifolia* (June 30 *Homoya and Hedge 4078*). On July 15 at least five plants of the nut-rush were seen in a remnant "barrens" environment near Leavenworth in Crawford County *(Homoya and Hedge 4201*).

Mikania scandens (L.) Willd. This vining member of the *Compositae* was found in a lowland forest along the Patoka River in Pike State Forest, Pike County, (August 26 *Hedge and Post s.n.*). While expected to occur in southwestern Indiana due to its proximity to populations in southern Illinois, it was previously known only from wetlands bordering the Kankakee River in northwestern Indiana. The Kankakee populations are no longer considered extant (1,9).

Infrequently Collected Species in Southern Indiana

Asplenium montanum Willd. Approximately thirty to fifty individuals of this mostly Appalachian spleenwort were observed growing on a south-facing sandstone cliff at Yellow Birch Ravine Nature Preserve in Crawford County (February 27 *J. and S. Aldrich, Homoya, Post, and C. and M. Ritter—photo voucher*).

Carex decomposita Muhl. Four new sites for this unusual semiepiphytic sedge were recorded for Indiana, all in Harrison County. Sites include a sinkhole swamp southeast of Palmyra (June 24, *Aldrich, Homoya, Post and Starcs s.n.*), a sinkhole swamp east of central Barren (July 15 *Hedge and Homoya 4196*), a sinkhole pond northwest of Laconia (July 28 *Homoya, Hedge, and Medley s.n.*), and a sinkhole swamp northeast of Palmyra (July 29 *Homoya, Hedge, and Medley s.n.*).

Carex eburnea Boott. The discovery of this sedge in Harrison County provides its second known occurrence in the state outside of the Lake Michigan dunes region. Numerous clumps of this plant form carpets over thin soil above and below both limestone and sandstone cliffs bordering the Ohio River at Harrison-Crawford State Forest (August 17 *Homoya, Babcock, Hedge, and Post s.n.*).

Carex gigantea Rudge. Extant populations of this southern sedge are currently known in Indiana only from a few sinkhole swamps in Harrison County. Plants were seen in swamps southeast of Palmyra (June 24 *Aldrich, Homoya, Post, and Starcs 4180*), and at three separate sites east of Central Barren (July 14 and 15 *Homoya and Hedge 4189*).

Dentaria multifida Muhl. While this species was reported from five counties by Deam (4), we currently know of only one extant site. Approximately twenty plants were observed in a mesic, forested rocky slope above the Vernon Fork of the Muscatatuck River at Crosley State Fish and Wildlife Area in Jennings County (April 30 *Homoya, Aldrich, Hedge, and Starcs s.n.*). Its small size and linear leaves easily distinguish this from the other *Dentaria* species with which it grows, including *D. laciniata, D. heterophylla,* and *D. diphylla.*

Dryopteris cristata (L.) Gray. The southernmost location for this mostly northern fern was Baxter's "Bog" in Monroe County until two new sites for it were found in Martin and Dubois Counties (April 21 *Homoya and Post 4006* and

May 24 *Homoya, Casebere, and Hedge s.n.*). Both new sites are acid seep springs dominated by *Osmunda cinnamomea* and *O. regalis. Sphagnum* is also present in parts of each seep.

Hottonia inflata Ell. Several hundred plants of American Featherfoil were found in a *Populus heterophylla—Taxodium distichum* swamp and a *Quercus lyrata* swamp two miles west of Hovey Lake State Fish and Wildlife Area in Posey County (May 6 *Homoya, Aldrich, Hedge, and Starcs 4123*). This is our only known extant site.

Oxydendrum arboreum (L.) DC. On February 28 fifteen to twenty trees were found in a sandstone ravine near Jubin Creek in Perry County *(Homoya, Aldrich, and C. Ritter s.n.*). This population of Sourwood is approximately twenty miles from the only other known site for it in the State (6).

Platanthera clavellata (Michaux) Luer. This diminutive orchid was found to be frequent on hummocks of *Sphagnum* and liverworts at the base of *Osmunda* ferns that occur in the acid seeps of Martin and Dubois Counties (May 24 *Homoya, Casebere, and Hedge*). Currently the nearest known sites in Indiana are from the northern tier of counties.

Platanthera flava (L.) Lindl. var. *flava*. A colony of fifty or more plants of this orchid were found in a floodplain forest along the Vernon Fork of the Muscatatuck River in Jackson County (April 29 *Hedge, Aldrich, Homoya, and Starcs s.n.*). Only seven plants of the group flowered; complete anthesis was achieved on July 14. This variety differs from the northern variety *herbiola* by its habitat preference, loose inflorescence, short bract length, quadrate shape of labellum, and later flowering date. The orchids were growing in a seasonally wet depression with *Saururus cernuus* and *Carex muskingumensis* and an overstory of *Liquidambar styraciflua* and *Quercus palustris.*

Poa paludigena Fern. and Wieg. A remarkable disjunct population of the Bog Bluegrass was rediscovered in a seep in Dubois County where Deam had found it in 1929 (May 24 *Hedge, Casebere, and Homoya 4155*). The site is the same one which harbors *Platanthera clavellata, Dryopteris cristata,* and *Itea virginica.*

Rhynchospora corniculata (Lam.) Gray. Two new sites for the Horned-Rush were found in northern Harrison County just east of Central Barren (July 14 *Homoya and Hedge 4229*). The plants were growing in sinkhole swamps with *Carex gigantea, C. lupulina,* and *Glyceria septentrionalis.*

Spiranthes tuberosa Raf. This infrequently collected orchid species (we have records of four previous statewide collections) is probably not as rare as once believed. We observed it flowering on two occasions,; at Yellow Birch Ravine Nature Preserve in Crawford County (August 18 *Hedge, Babcock, Homoya, Hutchison, and Post—photo voucher* and near Buck Creek in Harrison County (September 9 *Aldrich and Homoya—photo voucher*). Plants were growing with full exposure to the sun; in an old field and in an abandoned sandstone quarry. Associated species include *Viola pedata, Andropogon scoparius, Strophostyles umbellata, Bartonia virginica,* and *Lycopodium digitatum.*

Acknowledgments

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The Upland Sinkhole Swamps and Ponds of Harrison County, Indiana

Michael A. Homoya and Cloyce L. Hedge
Indiana Natural Heritage Program, Indiana Department of Natural Resources
Indianapolis, Indiana 46204

Introduction

Recent field exploration by members of the Indiana Natural Heritage Program has resulted in a better understanding of two little known natural communities in Indiana—the upland sinkhole swamp and pond. These natural communities, while studied extensively in surrounding states, have received little attention in Indiana. An inventory and study were initiated to provide a more complete understanding of these significant wetlands.

A sinkhole swamp is defined here as a forested wetland occupying a sinkhole depression, while a sinkhole pond is usually quite open and devoid of woody vegetation, also in a depression.

An inspection of Harrison County Soil Survey maps, aerial photos, and U.S.G.S. 7.5 minute topographic maps has revealed at least fourteen sinkhole swamp sites and a large number of sinkhole ponds, all in the Mitchell Plain region of the County. Those sites examined are quite small, varying from a quarter of an acre to five acres in size. They also vary considerably in natural quality and species diversity. A few sites yield a remarkable variety of Gulf Coastal Plain species, many currently known in Indiana from these sites only.

Methods

The U.S.G.S. 7.5 minute topographic maps and the S.C.S. Soil Survey aerial photos were inspected to locate potential sites for sinkhole swamps and ponds. While the topographic maps normally illustrate these environments well, they are not as revealing as the aerial photo base maps in the *Soil Survey of Harrison County, Indiana* (10). The photos, taken in winter, reveal areas of standing water in forests quite clearly. Those areas appear much darker on the photo than do the surrounding better drained sites. For example, some areas which simply appear green (as forest) with little or no contour on the topographic map were found by this method to contain fine quality sinkhole swamps.

Sites were field checked in the summer of 1982 to determine their natural community, degree of natural quality, and species composition. Specimen collections were made of rare or infrequently collected species to document their occurrence, while common species were recorded in field notes. The nomenclature used follows Fernald (3) with the exception of *Lorinseria*, which follows Cranfill (1). Voucher specimens will be deposited in the Deam Herbarium of Indiana University at Bloomington.

The Study Area

Sinkhole swamps and ponds are restricted in Indiana to the karst region, most of which occurs on the Mississippian limestones of the Mitchell Plain Subsection of the Highland Rim Section in the Interior Low Plateaus Physiographic Province (6,9). Our inventory concentrated on Harrison County, the southernmost county of the region. Exact locations for the sites are not given, but are on file with the Indiana Natural Heritage Program. The scattered local areas of karst

on Devonian limestone of Clark, Jefferson, and Jennings counties were not investigated.

The sinkhole swamps and ponds are not specifically associated with one soil type, but occur on a number of types, including Bartle, Bedford, Haymond, and Newark silt loams (10). However, a number of swamps in northern Harrison County do appear to be associated with one type—Bartle silt loam, which is described as an ". . . old alluvium in large basins in uplands." This soil type is found primarily in association with an unusually thick deposit of *terra rossa* soil which occurs as a one-to-two mile wide band running longitudinally through the middle of the County.

Water enters the swamps and ponds via small intermittant streams and from direct precipitation. There is usually no outlet due to the depressional nature of the sink, so seasonal fluctuation of water levels can sometimes be great. Depending on the size of the watershed, water levels can fluctuate from a few inches to several feet. During late summer many of the swamps have dried, though most retain some pockets of water. Most of the sinkhole ponds retain water the year around, due primarily to their concave shape which facilitates greater storage capacity than the more level swamp sites. For unknown reasons, the water level in a swamp may remain high for extended periods. All woody species and selected herbaceous species are subsequently killed, resulting in a significant loss of the swamp community. It may be only a temporary loss however, for most of the swamp species remain present on the periphery of the newly formed pond, ready to invade as suitable habitat becomes available. This process was noted where peat accumulation on the border of one pond was providing for the encroachment of woody species.

While sinkhole ponds are considered as distinct from sinkhole swamps in this paper, they appear to be essentially the same natural community type with the exception that ponds have deeper water. In fact, most undisturbed ponds are bordered by swamp and in turn, most swamps contain a pond in some portion.

History

The earliest written account of the occurrence of sinkhole wetlands can be found in the surveyor's field notes of the original land survey of the State in 1806 (12). At 67.5 chains north along the line between sections 32 and 33, T1S, R4E a surveyor noted: "A swamp 3 chains, 50 links wide full of scrubby water willows." A number of references were made by the surveyors concerning swamps, and most all mention willows. Interestingly, willow is not one of the major species of the swamps in this study, making one wonder if the sites have changed or if the surveyors were referring to something other than *Salix*.

Geologists have long recognized the sinkhole as a typical karst feature, but Clyde Mallott was especially skilled in noting and describing natural features associated with karst (6,7). He describes areas of poor surface drainage as solution pans, and when swampy, ". . . may be termed karst swamps or karst fens."

Botanist Charles Deam was interested in sinkhole ponds and kept a special list of plant species associated with them (2). It does not appear however that he was particularly impressed with them as natural areas. This is determined by his statement: "I have never seen any vegetation in them when they are located in deep woodland, doubtless because the decay of many leaves prevent it. Those in fields are disturbed more or less by stock which destroys much of their vegetation." With few exceptions our findings support his statements, for the great majority of the sinkhole ponds examined in this study are highly disturbed.

Deam's major contribution to our understanding of sinkhole swamps consists of collection data recorded for species he found at a site southeast of Palmyra. These data describe a swamp environment harboring such species as *Carex gigantea*, *Populus heterophylla*, *Rhynchospora corniculata*, and *Ranunculus pusillus*. During a visit to the site on June 24, members of the Heritage Program were able to reverify the existence of many of the species he found there in 1926. In addition, *Carex decomposita* and *Lorinseria areolata* were found, the latter new to the State.

After studying the geographic distribution of the species seen near Palmyra, it became apparent that the swamp had a high number of Gulf Coastal Plain species. This would have been viewed as merely interesting were it not for the knowledge that sinkhole environments in Kentucky and Missouri also contain notable assemblages of Gulf Coastal Plain species (11,5). Recognition that an association between sinkhole wetlands and coastal plain species is also true in Indiana inspired the intensive search that followed.

Plant Communities

The sinkhole swamps in Harrison County are characterized by the occurrence of water tolerant woody species, typically *Populus heterophylla*, *Liquidambar styraciflua*, *Nyssa sylvatica*, *Acer rubrum*, and *Quercus palustris*. *Populus heterophylla* appears to be the most tolerant of extended wetness, as it is usually found in the deepest water of the swamp. Here it often occurs with another woody species characteristic of these swamps, *Cephalanthus occidentalis*. At one site another shrub, *Itea virginica*, frequently appears as clonal thickets growing on tree roots, logs, stumps, and soil.

The herbaceous species appear to occur in distinct zones, responding to the degree and duration of wetness. Certain species appear to be restricted to the peripheral sections of the swamp where standing water is only a winter and spring phenomenon. Species seen most frequently in this zone include *Carex gigantea*, *c. lupulina*, *C. intumescens*, *C. crus-corvi*, *C. grayii*, *Habenaria peramoena*, *Glyceria striata*, *Galium tinctorium*, *Lorinseria areolata*, *Cinna arundinacea*, *Leersia oryzoides*, *Iris virginica*, *Impatiens biflora*, *Rhynchospora corniculata*, *Triadenum walteri*, and *Sphagnum* sp. In the zone where water is found standing for most if not all months of some wet years, another set of species can be observed. These include *Bidens cernua*, *Polygonum hydropiperoides*, *Glyceria septentrionalis*, *Proserpinaca palustris*, *Peltandra virginica*, *Ludwigia palustris*, *Lemna* sp., *Wolffia* sp., and *Spirodela* sp. Here also *Rosa palustris*, *Lycopus rubellus*, *Triadenum walteri*, and *Carex decomposita* are often found growing on floating logs, stumps, and bases of living trees, most often *Populus heterophylla*.

Three exceptionally fine examples of a sinkhole pond community can be found just northwest of Laconia in southern Harrison County. The ponds occur no more than a few hundred yards from one another, yet each is unique. The first pond, an acre in size, is best characterized by a three inch thick mat of organic detritus which floats on one to two feet of water. This mat harbors such species as *Dulichium arundinaceum*, *Carex comosa*, *Leersia oryzoides*, *Utricularia gibba*, and *Bidens* spp. On the border of the pond *Nuphar advena*, *Salix nigra*, *Typha latifolia*, *Hibiscus palustris*, and *Cephalanthus occidentalis* occur. This border grades outwardly to a *Quercus palustris* swamp.

The second pond, slightly larger than the first, is dominated by a nearly impenetrable thicket of *Decodon verticillatus*. Within this thicket one small opening occurs where *Nuphar advena* and *Carex comosa* predominate. On the hum-

mocks formed at the base of *Decodon* large growths of *Carex decomposita* can be found. On the periphery a variety of species can be found, including *Alisma subcordatum, Impatiens biflora, Rosa palustris, Eupatorium perfoliatum, Glyceria striata, G. septentrionalis, Triadenum walteri, Nuphar advena, Cephalanthus occidentalis, Sparganium androcladum, Dulichium arundinaceum,* and *Carex comosa.* This peripheral area grades into swamp, where *Liquidambar styraciflua, Quercus palustris, Q. bicolor, Nyssa sylvatica,* and *Populus heterophylla* occur.

The third pond, about five acres in size, is mostly an expanse of marsh surrounding a small, open body of water. Plants in the shallow section include *Dulichium arundinaceum, Proserpinaca palustris, Alisma subcordatum,* and *Eleocharis* spp. while in the deeper section *Sparganium androcladum, Typha latifolia, Scirpus validus,* and *Nuphas advena* can be found.

Results and Discussion

The sinkhole wetlands in Indiana appear to be similar to those in Missouri and Kentucky. Steyermark (11) and Haefner (4) describe a number of species associated with sinkhole wetlands in the Ozarks of Missouri. Species they list as characteristic of sinkhole wetlands that are also in such environments in Indiana include *Carex decomposita, C. comosa, Dulichium arundinaceum, Glyceria acutiflora, Decodon verticillatus, Galium tinctorium, Cephalanthus occidentalis, Rosa palustris, Isoetes englemannii, Lycopus rubellus, Triadenum walteri,* and *Utricularia gibba.* The sinkhole wetlands in Kentucky also contain species known to occur in similar Indiana sites. Some of these are *Isoetes engelmannii, Carex crus-corvi, Rhynchospora corniculata, Quercus palustris, Polygonum hydropiperoides, Ranunculus pusillus, Liquidambar styraciflua, Triadenum walteri, Ludwigia palustris, Bacopa rotundifolia,* and *Itea virginica* (5,8).

In both Missouri and Kentucky a number of species found in sinkhole wetlands have Gulf Coastal Plain affinities. This is true in Indiana also, as indicated by the presence of *Carex gigantea, C. decomposita, Triadenum walteri, Lorinseria areolata, Rhynchospora corniculata, Itea virginica,* and *Ranunculus pusillus.* Coastal Plain species in sinkhole wetlands are suggested by Steyermark (11) and Hannan (5) to be relicts of the Teriary Period, when the Ozark and Mississippian Plateaus were low, swampy peneplains and the climate subtropical. The sinkhole wetlands are just remnants of the once vast swamp, now providing refugia for these relict species. This hypothesis may also explain the occurrence of Coastal Plain species in Indiana sinkhole wetlands. It is not however within the scope of this paper to provide an hypothesis or to confront the problems of plant geography. However, the occurrence of these species with Coastal Plain affinities in the karst regions of the three states does tend to indicate a common hypothesis concerning their past and present distribution.

Human disturbance has significantly reduced the sinkhole wetlands to only a few good examples of their former character. Timber harvest, drainage, and conversion to cattle ponds have all but eliminated them as natural areas. Of the fourteen sinkhole swamps identified in this study, only four have some semblance of natural quality with only three sinkhole ponds exhibiting little or no disturbance. It appears that they, like so many other natural features in the State, are being eliminated at an alarming rate.

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The Arthur Herbarium Centennial: 100 Years of Uredinology in Indiana and the Great Lakes Region

JOHN W. MCCAIN and JOE F. HENNEN
The Arthur Herbarium, Department of Botany and Plant Pathology
Purdue University, West Lafayette, Indiana 47907

In January, 1883, Dr. J. C. Arthur published his first manuscript on uredinology, the study of the taxonomy and biology of the rust fungi, the Uredinales. This we take as the starting date for the Arthur Herbarium. Because the Arthur Herbarium (PUR), now at Purdue University in West Lafayette, Indiana, has now for 100 years been the major site in the United States, and possibly the world, for taxonomic studies of these important plant pathogens, a review of the history of Indiana Uredinology is in order. Parts of Dr. Arthur's story have been published before (11, 16, 17, 19, 24, 30), so new information was sought for this report.

The Arthur Herbarium is in its centennial year, but rust collecting in Indiana began more than 100 years ago. Jackson (23) credited the first explicit report of a rust fungus in Indiana to Dr. John M. Coulter in 1876, who found a rust on *Lespedeza violacea* (L.) Pers. (bush clover: Leguminosae), presumably near Hanover. Because there was then no Arthur Herbarium, Coulter sent the rust specimen to Dr. Charles H. Peck, New York State Museum at Albany, for identification. Peck indentified the fungus as *Uromyces lespedezae-procumbentis* (Schw.) Curtis, then called *U. lespedezae-violaceae* Schw., but added,

> " . . . inasmuch as the name is objectionable both because of its compound character and because of it implied limitation of the habitat of the fungus, which occurs on the leaves of all of our species of *Lespedeza*, it seems best to . . . change . . . the name of this fungus (14)."

Therefore, Peck simply shortened it to *Uromyces lespedezae* Peck, a savings of four syllables and twelve letters—a noble, but unfortunately an illegal, change under the International Code of Botanical Nomenclature.

John M. Coulter is known to Indiana botanists as a professor of botany and as the editor of the *Botanical Gazette*. He was also the brother of Stanley M. Coulter, early curator of the vascular plant herbarium at Purdue University. Perhaps it is less well known why J. M. Coulter became a botanist. Although Coulter studied botany in college, his main interest was geology—and Latin (31). His first scientific position was as an assistant on a U.S. Geological Survey of Colorado, Utah, Idaho, Montana, and Wyoming. Every evening his companions socialized. Because of his inability to play cards, Coulter sought something else to do and became interested in the plants of the "Far West (35)."

There is a less exact reference to a rust disease in Indiana two years prior to Coulter's report. A rust was destroying the foliage of blackberries in Henry and Wayne counties from 1872-1874 (22). The reporter stated, "It is evidently a contagious trouble. I think it of cryptogamous origin." No Latin name or further description was included, but it probably was the orange rust of *Rubus*, caused by *Gymnoconia peckiana* (Howe) Trott.

Another nebulous early report is of a specimen labeled "*Aecidium dircatatum* Ind." Arthur and Bisby (9) interpreted this notation to mean that it "must have

been collected" by the Moravian missionary, Lewis David von Schweinitz, during a visit to Hope, Indiana, in 1831. The fungus, now known as *A. hydnoideum* Berk. & Curt., was collected on *Dirca palustris* L. (Thymeleaceae). This specimen was not available to us in time for verification of this report.

The first compilation of Indiana rust fungi appeared in the Indiana Academy of Science proceedings for 1893 (36). The tally included 7 holomorphic genera, 4 anamorphic genera, and 88 species. Several of the *Aecidium* anamorphs are now connected to teliomorphs in such genera as *Gymnoconia*, *Pileolaria*, and *Tranzschelia*.

Thus, Indiana uredinology dates to 1831, 1872, or 1876, depending on the validity of the blackberry and *Dirca* rust reports. How does this compare to other Great Lakes states or to the rest of North America? The first American fungal studies were those published by von Schweinitz in 1831 in Pennsylvania (1). These could hardly have been done earlier, for Schweinitz still had to combat "the prevalent notion of the times that fungi were mutations or transformations usually from parts of higher organisms" and, therefore, not suited for true classification (7).

In Ohio, the earliest literature report of a rust fungus appears to be of *Uromyces ari-triphylli* (Schw.) Seeler on *Arisaema dracontium* (L.) Schott. (green dragon: Araceae) near Toledo in 1877 (21). Burrill reported *Aecidium physalidis* Burr. *Sp. nov.* on *Physalis viscosa* L. (=*P. heterophylla* Nees., groundcherry: Solanaceae) from Urbana, Illinois, in 1879 (13). This anamorph may be associated with the species *Puccinia kansensis* Ell. & Barth. on *Buchloë* (buffalo grass: Graminae) (10). The earliest rust collection on record in the Arthur Herbarium for the state of Michigan is the type specimen of *Aecidium polygalinum* Peck on *Polygala senega* L. (Seneca snakeroot: Polygalaceae). Now known to be an anamorph of *Puccinia andropogonis* Schw. (on *Andropogon* spp., bluestem grasses: Gramineae) (29), it was collected near Ann Arbor in 1878. One of the earliest lists of rust species in Michigan was by Lee Bonar (12), later a distinguished California mycologist but then working as a graduate teaching assistant at the University of Michigan for a salary of $300.00 per year (34).

The oldest specimens of rust fungi to be found in herbaria usually were collected incidentally as higher plant taxonomists were sampling the host plants of the rusts. So it was with the oldest PUR collection (PUR-F18269). This fungus, *Puccinia meyeri-alberti* P. Magn. (27), caused a tiny leafspot on a specimen of *Berberis ilicifolia* Forst. (Berberidaceae) collected in Tierra del Fuego in January, 1769, by Sir Joseph Banks and Dr. Daniel Carl Solander during the first voyage of Captain James Cook. The expedition party was trapped overnight in a blizzard on a mountainside after collecting this plant. Two black servants, Thomas Richmond and George Dorlton, froze to death (32) but are memorialized by the Arthur Herbarium's oldest specimen.

Thus, uredinology began in several of the Great Lakes states at about the same time, but Indiana is the state where rust taxonomy flourished. This can be credited to the work of one man: Joseph Charles Arthur. Arthur showed early promise as a botanist. He passed botany exams given by his Iowa State University professor, Dr. Charles E. Bessey, by being "able to give the Latin names of the required fifty specimens by the shadow seen through the mounting paper when the sheets of dried plants were held at the window with the backs turned toward" the students (31). Dr. Arthur began his studies of rust fungi over 100 years ago, in 1877, when Dr. Bessey acquired a collection of unidentified rusts from the Curtis Herbarium at Harvard. Among these was the subject of Arthur's first publication on rust fungi (30), the same species Coulter found in Indiana in 1876, *Uromyces lespedezae-procumbentis*. This publication (3) began the first cen-

tury of sharing with the world knowledge gained from what became Arthur's herbarium.

Arthur's second rust publication (5) soon followed but was published obscurely because, in Arthur's words (7),

> " . . . it was then held and long afterward by Dr. Asa Gray and concurred in by Dr. Farlow, that for the good of American science no naming of phanaerogamic or cryptogamic plants, particularly the latter, should be undertaken outside the precincts of Harvard University, because in no other place was there to be found adequate material for comparison."

W. G. Farlow then held the Chair of Cryptogamic Botany at Harvard. Because this work was done while Arthur was an instructor at the University of Wisconsin, Farlow refused to acknowledge it, even though he and Arthur were good friends. However, Arthur's visit to Farlow's laboratory in 1884 led Arthur to specialize in the rust fungi. Seeing that Farlow had a backlog of unexamined specimens (including some of Arthur's own), Arthur decided that Farlow could not manage the task alone. In a letter to Bessey, Arthur announced that he had "concluded to look into the life and habits of some of the plants that cause diseases of cultivated crops, such as the rusts etc. (31)." Dr. Arthur came to Purdue a few years later and, through his work, now at least one place other than Harvard contains adequate rust material for comparison of specimens.

In his second rust manuscript (5), Arthur's future research direction is evident. Perhaps this course was explained best in a review by Coulter (15):

> " . . . if ever the study of the lower cryptogams ceases to be a bugbear, it will largely be due to just such workers as Prof. Arthur. The novel part about the present paper is that it attempts to group under each species all the three stages in its life history. Of course this is the thing to do, and it has only been because of our ignorance of the true relationship among these scattered phases, that it has not long since been done. A careful set of cultures is what is needed to unravel the snarl. In the meantime it has been customary to base species upon the characters of one or two phases, and to distribute the phases under separate genera, and Prof. Arthur shows considerable boldness in cutting loose from the old models, and attempting a natural grouping."

This article contains the first of Arthur's numerous published species of Uredinales, *Uromyces acuminatus* Arth. on *Spartina pectinata* Link. (prairie cordgrass: Gramineae), and the new variety *U. euphorbiae* Cke. & Peck in Peck var. *minor* Arth. (on *Euphorbia marginata* Pursh: Euphorbiaceae), a taxon we do not still accept. Surprisingly, Arthur's first named taxon was not a rust fungus but a new variety of walking fern, *Camptosorus rhizophyllus* Link var. *intermedius* Arth. (4), published one month before his first named rust species.

Arthur came to Purdue University in 1887 as Professor of Botany, became botanist in the Purdue Agricultural Experiment Station one year later, and remained at Purdue even after his retirement in 1915. His friendship with J. M. Coulter and with C. R. Barnes, Arthur's predecessor as botany teacher at Purdue, led to collaboration in the textbook *Handbook of Plant Dissection* (8), which became a standard that students used to learn their ABC's (Arthur, Barnes, and Coulter) of plant anatomy. Note that the ABC publication predated that famous alphabetically-authored cosmology paper of Alpher, Bethe, and Gamow (2) by 62 years.

Arthur seems in some ways like a part of the distant past, a picture on the wall, but 100 years is not so long. Indeed, some of his ideas sound like 1982. For example, consider the following excerpt from another unpublished Arthur note (7), on file in the Arthur Herbarium:

> " . . . Those who fatten at the public feeding trough, have at times been transformed from statesmen into pork-barrel politicians, but it would not be expected that a parallel could be found in the non-monetary field of botany. But something very like it has happened. Among the Washington botanists, quite an army at present, one can expect little cooperative assistance.
>
> "My acceptance of the task of listing and describing the rusts of North America began before anyone of those who now guard their discoveries among the rusts with the methods of a secret service man had any flickering of interest in the subject, so long ago that the New York Botanical Garden, which has fostered the enteprise, was only in the heads of its promoters, and the extensive grounds were an unopened, rocky pasture.
>
> "I undertook the work with the conviction . . . that it was to be for the benefit of science in general . . . I received no financial aid from any source whatever. I bought all books needed in my works from my slender salary as a teacher, a salary so small that I felt ashamed to name the amount, even to a friend . . . My work was done at odd hours, often at the sacrifice of social and recreative periods, and even of normal hours of sleep."

The work mentioned in this quotation was the portion of the North American Flora devoted to the rust fungi (6), which had been assigned to Arthur in 1885 (31). Although we do not know when or why these paragraphs were written, some of Arthur's personality shows through in this quote. For example, there is that pride in his work which led him to pack up the entire herbarium and move it to his home during a disagreement with Purdue University over ownership of the herbarium (17). When the debate was resolved, on 05 August 1918, and the herbarium returned to the campus, it was officially dedicated as the "Arthur Herbarium."

Arthur, though he retired in 1915, stayed active as curator of the herbarium until about 1938, when he was 88 years of age. Thus, not only are the PUR specimens well-preserved, but so, apparently, are the Arthur Herbarium researchers. One of Arthur's early assistants, Dr. Frank D. Kern, published his last book (26) at 90. Arthur's successor, Dr. George B. Cummins, named over 600 new taxa or combinations in a 33 year career as curator of the Arthur Herbarium and best softball pitcher in the Purdue University School of Agriculture. However, Cummins has been even more prolific as a writer following his formal retirement, and is now finishing his sixth book since leaving Purdue. Cummins, who signs his letters "the burned-out botanist," is 78 years old but not yet rusty. The following quote (18) is representative of Cummins' trademark dry wit:

> " . . . The abbreviation of Purdue University Retirees (PUR) is antedated by some years by PUR, the official international designation for the Arthur Herbarium . . . The two PUR's have something in common. The PUR (Arthur Herbarium) is a repository of about 80,000 specimens, many quite old. Purdue University Retirees probably do not number so many, but the rest of the comparison has some validity. And I am in a position to know."

Finally, still actively working in the Arthur Herbarium today is Ibra L. Conners,

retired former Curator of the Canadian National Mycological Herbarium in Ottawa, Ontario, who celebrated his 88th birthday on 05 July 1982.

This has been a brief review of Indiana uredinology and of the Arthur Herbarium on the occasion of its centennial celebration. What is the status and significance of the PUR today? Specimen holdings now approach 100,000 (Table 1). No up-to-date compilation of Indiana rust fungi is available, but a recent tally

TABLE 1. *Specimen and record holdings of the Arthur Herbarium (PUR), October 1982* (estimated figures)

86,000	accessioned rust fungus specimens
10,000	new rust fungus specimens under study
3,000	type specimens of rust fungi
5,000	specimens of other fungi for teaching use
120	teliomorphic genera of rust fungi (of 120 known in the world)
9	of the 10 anamorphic genera of rust fungi
4,000	species of rust fungi (of 6,000 known in the world)
1,000	genera of host plants
5,000	species of host plants
4,000	photomicrographs, camera lucida and free-hand drawings, annotation notes (including spore measurements)
23,000	entry bibliographic file
6,000	reprints from world literature
21	U.S. states-sources of correspondence or specimens exchanged with PUR in last two years
26	foreign countries-sources of contacts within last two years

of Michigan rusts (McCain, unpublished manuscript) included 29 genera, 171 species, and 6 varieties, a notable increase over Underwood's 1893 list of 88 Indiana rust fungi. The herbarium is a valuable source of plant distribution records, e.g., some of the citations in Deam (20) came from PUR records. The herbarium is also a reference for identification of species, such as the first Indiana record of *Viburnum* rust, *Coleosporium viburni* Arth., which was collected on *V. lentago* L. (nannyberry: Caprifoliaceae) from South Bend, IN, on 3 September 1982, by B. Cummings (PUR 66499). Another use of PUR records is for biogeographic studies (28). There is still no explanation for such extraordinarily disjunct species as *Cerotelium tanakae* Ito, which occurs on *Amphicarpa* (hog-peanut: Leguminosae) in Japan and in Brown County, Indiana, but apparently nowhere else. The rust was collected again in 1982 at the previous location in a ravine near the Abe Martin Lodge in Brown County State Park. However, in a survey of the host in other nearby locales, P. Buriticá and J. F. Hennen failed to locate any additional rust specimens.

In short, as the Arthur Herbarium begins its second century, it continues to be a haven for taxonomists, "those sainted professionals who have taken on the ostensibly boring task of naming new species and untangling the web of synonyms applies to old ones (33)," morphologists and ultrastructurists, those who study by the light of the flickering cathode ray tube, and phylogenists, those who seek to answer the old question: which came first—*Sempervivum tectorum* L. or *Solanum melongena* L.?

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SCIENCE EDUCATION

Chairman: PATRICIA A. ZECK
Northwestern High School
Kokomo, Indiana 46901

Chairman-Elect: STANLEY S. SHIMER
Science Teaching Center
Indiana State University, Terre Haute, Indiana 47809

ABSTRACTS

Indiana Public School Enrollment Trends in High School Science Courses (1968-1981). JERRY M. COLGLAZIER, Science Consultant, Division of Curriculum, Indiana Department of Public Instruction, Indianapolis, Indiana 46204.—— Data annually collected and totaled by the Indiana Department of Public Instruction regarding Indiana public school enrollments in high school courses were tabulated and reviewed to identify enrollment trends in science courses. Although the number of students enrolled in high school science courses increased in the early seventies, this increase did not keep up with the increase in the early seventies, this increase did not keep up with the increase in total high school membership, in fact the percent of students enrolled in science, particularly second, third and four year science courses, declined up through the 1973-74 school year. From the 1975-76 school year through the 1979-80 school year, the percentage of students enrolled in science showed a very slight upward trend; however, the percentage enrolled in advanced courses remained nearly constant. During the last two years (1980-81 and 1981-82), the percentage of students enrolling in science has turned perceivably upward. Increased enrollment in biology and chemistry account for most of this increase although a slight increase is detectable in the physics enrollment data. Analysis of class data seem to indicate that beginning with the graduating class of 1983, Indiana public school students began enrolling in science in perceivably larger percentages. It will be interesting to see if: (1) this trend continues and (2) if physics enrollment percentages have increased this school year. In the fall of 1981, 49.8% of Indiana's public high school students were enrolled in a science course compared to a low of 46.8% in the fall of 1973. The equivalent of 74.0% of the 1981 freshmen were enrolled in biology, 24.2% of the 1979 freshmen were taking chemistry and 9.7% of the 1978 freshmen had elected to take physics.

Results of an Inservice Needs Assessment for Improving Science Instruction in Indiana Schools. JERRY M. COLGLAZIER, Science Consultant, Division of Curriculum, Indiana Department of Public Instruction, Indianapolis, Indiana 46204.—— A random sample of Indiana public school elementary teachers, of middle school science teachers and of high school science teachers and all science supervisors were sent a questionnaire in April, 1982. In addition to some demographic data, the questionnaire asked respondents to indicate how valuable they considered each of 29 categories of inservice programs and 14 possible other support services. They were also asked to rate possible inservice delivery arrangements. The return rate was about 30.0% for all teacher groups and 88.0% for supervisors. The data was processed using the Statistical Package for the Social Sciences.

Science Requirements for Indiana Schools: Past, Present and Future. JERRY M. COLGLAZIER, Science Consultant, Division of Curriculum, Indiana Department of Public Instruction, Indianapolis, Indiana 46204.——For many years, the rules of the Commission on General Education, Indiana State Board of Education, relating to the science curriculum, in fact the total curriculum, remained constant. These offered only suggested guidelines for the content areas to be included in grades one to eight while requiring one year of laboratory science for high school graduation. The list of science courses acceptable to meet this requirement was altered from time to time between 1937 and 1978; however, these alterations were always somewhat limited in scope. In February, 1978, the Commission promulgated a new set of rules (actually a section of Indiana Administrative Code) which became effective in June, 1978. In terms of curriculum, the main changes from previous rules were that of specifying minimum time allocations for each subject in grades kindergarten through eight. In terms of science, they required that 15.0% of the kindergarten program be devoted to social living and environmental experiences; 5% of the time in grades 1-3 be devoted to science/health/safety education; and 10% of the grades 4-8 be devoted to science. During 1982, the Commission has been considering a revision of the curriculum sections of the Indiana Administrative Code. Proposed revisions would separate science and health/safety education in grades 1-3, allocating 5.0% of the instructional time to each one; increase the 10.0% requirement to 12.0% in the middle school grades; and greatly expand the list of acceptable high school science courses.

Solving Moles Problems: Strategies and Errors. DOROTHY GABEL, Department of Science and Environmental Education, Indiana University, Bloomington, Indiana 47405.——During the 1979-80 school year 74 high school students in south central Indiana from seven high schools were taped while they solved three chemistry mole problems aloud. Problems became increasingly more difficult beginning with one step problems and culminating in three step transfer problems. Before students began solving the mole problems, their background of prerequisite knowledge necessary for solving the problems was assessed by asking them a series of questions on the concept itself.

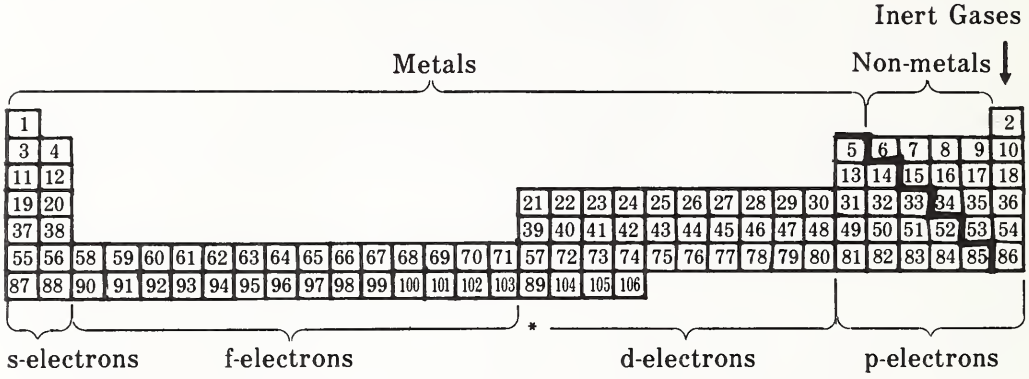
Students' responses were coded and classified according to the type of errors made. Tapes were further coded to determine the general problem solving strategies used according to Polya's heuristics. Data were analyzed using nonparametric statistics.

Results of the study indicate that students' major difficulty in solving mole probelms is lack of understanding of the mole concept. Specific errors that were identified could be classified as (1) using the wrong operation but understanding the concept; (2) lack of understanding of the mole itself, e.g. confusing moles with mass, volume and molecules; (3) using the wrong conversion factors and (4) failure to use moles in the problems. Students generally solved problems using only algorithms and only rarely check to determine the reasonableness of their answers.

A Teaching Tool for Introducing Elements: The Expanded Periodic Table. LINDA HAMRICK, The Canterbury School, Fort Wayne, Indiana 46807.——In 16 years of teaching general science, introductory utilization of the Periodic Table of Elements invariably brings the question, "What about the elements that are 'left over' across the bottom?" Although chemists certainly have a firm grasp of the place the f-electron Rare Earth Elements occupy in the overall scheme of things, for students encountering the table initially, the positioning of the Lanthanides and Actinides can be a source of confusion. One answer which the author developed and found

helpful over 8 years of use is an Expanded Periodic Table which fits the Rare Earth Elements into a continuous array with the rest.

EXPANDED PERIODIC TABLE



Atomic numbers 51 and 89, (asterisk in the table above), dropping back and adding the first d-electron in the Transition Elements is the only discontinuity, (and a minor one compared to the student-view of an entire group of 14 elements being "forgotten and added on later" which is the typical perception). The f-electrons then fill in neatly, leap over 51 and 89 (already added), and proceed smoothly across the d and p-elements.

Although there is not enough room in the squares for detailed information, the presence of numbers does provide the required identification, while maintaining an introductory simplicity which is highly conducive to understanding in the newer student. Later, when the pupil has a feel for the inherent unity of the elemental relationships, the idea that the size of paper in combination with the amount of information to be squeezed into each square, necessitates "pulling out" the Lanthanides and Actinides, and placing them at the bottom is explained. It is not, of course, proposed that this version replace the current format of the Periodic Table, but rather be utilized as an initial teaching tool for introducing a clear rationale behind its structure.

T.I.P.—An Intensive Science Tutorial. G. C. KYKER, JR. and F. A. GUTHRIE, Rose-Hulman Institute of Technology, Terre Haute, Indiana 47803, and W. A. Deutschman, Oregon Institute of Technology, Klamath Falls, Oregon.——The Iceberg Project (T.I.P.) is a two-week summer program for exceptional students who will be high school seniors in the upcoming academic year. It has been operated by Rose-Hulman for the past three summers. A research-oriented program rather than a "course" as such, its goal is to provide the student with an experience of scientific investigation as it actually takes place. The special characteristic of this program is the intensive collaboration of a faculty member with a small group of no more than six students. Students spend well over half the time in their small group, and can deal with their particular research poblem in considerable depth. Representative topics have included neutron activation analysis, holography, organic synthesis, and games theory. Other features of the program include an introduction to digital computer use, both for scientific programming and word processing; production, entirely by the students, of a collaborative research report; and evening programs on scientific topics of general interest. These and other aspects of the program will be discussed, and our fist three years' experience with it evaluated.

Chemistry Laboratory Experiments Encouraging Abstract Reasoning: Their Use in the High School. JOHN A. RICKETTS, Department of Chemistry, DePauw University, Greencastle, Indiana 46135 and LUCY BROOKS, Western Boone Junior-Senior High School, Crawfordsville, Indiana 47933.—A set of chemistry laboratory experiments will be described. Their experimental design is based upon the Piagetian model for cognitive development; each experiment is constructed as a Learning Cycle, as described by Robert Karplus (SCIS, Teachers' Handbook (1974), Lawrence Hall of Science, Berkeley, California). A salient feature of the Learning Cycle is its devision into three distinct parts, Exploration, Invention, and Application. In these laboratory experiments the three parts are separate experimentally; however, they are connected logically. The observations and conclusions from Exploration are necessary to understand Invention; the conclusions reached in Exploration and Invention are necessary to formulating and understanding the Application.

Abstract and analytical reasoning skills are essential to the understanding and mastery of all academic disciplines. Little or no attention is given to the student within his/her high school experience to strengthen reasoning abilities; consequently, the initial college experience can be an academic disaster—especially in those disciplines that use the hypothetico-deductive method coupled with quantitative reasoning. With the Learning Cycle the science laboratory can be constructed to introduce the techniques of the science and simultaneously develop reasoning abilities. Four specific chemistry Learning Cycles, (1) The Chemical Dilemma, (2) Indictive Reasoning from Chemical Experiment, (3) The Plot Thickens, and (4) The Great Titration Mystery, were tested within a high school chemistry class of twelve students at Western Boone Junior-Senior High School.

The reasoning abilities of the students were assessed before and after performing the laboratory using puzzles requiring proportional, probabalistic, and combinatorial reasoning abilities. Within the group of twelve students, seven exhibited marked improvement in their reasoning abilities. Two who were initially categorized as concrete operational remained concrete operational; the three who were initially classified as formal operational remained formal operational.

Teaching to Overcome Personal Preference of Topics in Zoology. PATRICIA A. ZECK, Northwestern High School, Kokomo, Indiana 46901.——To determine if teacher predilection of topics in zoology had any effect on student preference of the same topics, the students and teacher independently ranked eight topics according to preference. Seventy-six high school students in the course chose seminar, genetics, and vertebrate anatomy and physiology as their favorites. The teacher picked ecology, genetics, and seminar. Their next cluster of choices was embryology and invertebrate anatomy and physiology. Their last group was ecology, evolution, and science projects. Seminars (*Proc Ind Acad* 86:416) were first because of their unique diversity. Science projects were last because of the amount of work required. Students ranked anatomy, physiology, and embryology higher than the teacher. Consequently, it is possible to convey successfully enthusiasm for a topic that is not necessarily the teacher's favorite.

The following teaching methods were used to achieve this success:

1. Approach every topic with a positive attitude.
2. Be "over prepared" especially on difficult material.
3. Use a variety of media such as handouts to match overhead transparencies, movies, written outlines of each lecture-discussion, vocabulary lists, specific quantities of lab work per session.

4. Stress applications of each topic to careers and college education.
5. Remember that teacher enthusiasm and patience stimulate students' learning and enjoyment.

Enrichment Programs for Secondary Science Students

WALTER A. CORY, JR.
Coordinator for School Sciences
Indiana University
Bloomington, Indiana 47405

A study in 1975 by the American Institute of Research dealt with the follow-up of a sample of persons who participated in Project Talent while attending high school in 1960. They were asked what they remembered, what they used, and what they thought was worth learning in their high school curriculum. With very few exceptions, these individuals, then in their 30's, replied that the only things they remembered and thought worthwhile in their high school courses were those that they used in their work (e.g., mathematics in engineering, English composition in newspaper reporting, home economics in the operation of a business) or those other subjects in which they were deeply interested. Science was listed as a subject of such deep interest by very few who were not then engaged in occupations that utilize science (8).

How can we improve the understanding of science by the majority of our students who do not become scientists or engineers? To do this, I believe we must balance *content* and *process* in our classes, but we must not just *tell* students about science. We must lead them through simple experiments that demonstrate logical thought and give them more and more responsibility for their learning.

Eventually, we should involve most students in independent scientific research with an emphasis upon written and oral presentation of their findings. There are many good books and articles available to assist teachers and students who wish to improve in this area (2).

Students must recognize that learning is a life-long activity, not just something to do to mark time until one reaches age 16, or 22 and then terminates formal education. Our students must also understand that those who pursue a career in a scientific or technical field can expect to devote more than 40 hours to work each week. Hopefully, they will enjoy most of what they will do in that field.

Today, we face many very difficult problems in education at all levels in these United States. Many of us who have worked hard to do a good job of educating our youth for many years have become discouraged and disillusioned by the paucity of interest demonstrated by our society and even our students in our chosen field of education. This is especially true in science education. We need to attract bright young students to teaching science, mathematics and computer science. Such service may be for just a few years, or it may be for many decades. We need such involvement for as long as we can get it! Teaching is a very important job!

It is very difficult to work hard to do a good job of teaching under the conditions which exist in many, if not most, of our schools today. But I want you to keep trying! Don't become disillusioned! There are many organizations willing and anxious to help you and your students to learn about the products and the processes of science. The Indiana Academy of Science is one of these; the Hoosier Association of Science Teachers is another.

There are signs that the lot of science education will improve very soon, but I cannot say how much improvement we can expect. I am very encouraged by what is beginning to happen around the country. In Indiana, we have many problems, but we also have many good things working for us. I consider this

Academy a sleeping giant in this regard. We have a great wealth of talent in our ranks, but we need to become more involved in education at both the state and national levels if we are to meet some of our pressing needs in Indiana.

Let me describe now some of the programs in which you and your students might become involved this year or next year. I think you will find such involvement very rewarding and very stimulating. Even if it involves begging or granting extra credit to your students, *please* give some of these programs a try in your school (3).

WESTINGHOUSE SCIENCE TALENT SEARCH—NATIONAL COMPETITION (for High School *SENIORS ONLY*). Science scholarships totaling more than $89,000 are provided for 40 students selected to attend the Science Talent Search in Washington, D.C., each spring. The Search fosters the education of potential scientists and focuses attention on the need for perfecting scientific research skills and knowledge. Honors Group status, earned by the top 300 students who complete entries, is evidence of high science ability and interest which is taken into consideration by colleges and universities in decisions regarding admissions and granting of scholarships. Studies involving vertebrate animals are usually not permitted. In 1981-82, only 950 students entered this competition. The deadline for submitting materials is December 15. Teachers should request application materials and the brochure, "Science Talent", which contains additional information, including the name and address of each state director from:

Mrs. Dorothy Schriver
Science Service
1719 N Street, N.W.
Washington, D.C. 20036

SCIENCE TALENT SEARCH—STATE COMPETITIONS. In Indiana, each entrant must submit a data form along with *two* copies of his or her research report. If selected as a finalist, he or she will be interviewed by a panel of research scientists which chooses the state winners. The Indiana competition is open to both juniors and seniors, with emphasis upon the excellence and completeness of the student's investigation. Studies which involve vertebrate animals are permitted only if humane and scientifically acceptable procedures are employed. Although group entries are acceptable, individual entries are strongly recommended. The deadline for submitting materials is December 15, and the Honors Weekend for finalists is held in March at the Medical Center Union Building in Indianapolis. Currently, two scholarships in the amount of $1,000 each from Kappa Kappa Kappa Sorority are available each year for the top male and female entrants. In Indiana, write:

Director, Indiana Science Talent Search
Education Building 253, Indiana University
Bloomington, Indiana 47405

In Table 1, winners in the Westinghouse Science Talent Search from 1942-1982 are tabulated by state and territory (6). Residents of New York, Illinois and California have been very successful in gaining recognition for themselves and their schools. Students from Indiana have not fared nearly so well, but they have placed Indiana in twelfth place with a total of 38 winners in the WSTS. Considering the population of Indiana and of those states ranked number 1-11, Indiana residents have done very well. However, we have the potential to do much better.

TABLE 1. *WSTS Winners By State/Territory (1942-82)*

State	Totals	Rank	State	Totals	Rank
New York	502	1	Iowa	10	29
Illinois	223	2	Hawaii	9	30
California	220	3	Kansas	9	30
Pennsylvania	81	4	Montana	9	30
New Jersey	66	5	Alabama	9	30
Ohio	66	5	Kentucky	7	34
Massachusetts	54	7	Louisiana	7	34
Florida	51	8	Rhode Island	7	34
Virginia	40	9	Maine	6	37
Texas	40	9	South Carolina	6	37
Wisconsin	40	9	Idaho	5	39
Indiana	38	12	New Mexico	5	39
Maryland	34	13	Mississippi	4	41
Oregon	25	14	North Carolina	4	41
Connecticut	25	14	South Dakota	4	41
Arizona	21	16	Wyoming	4	41
Oklahoma	20	17	Alaska	3	45
Minnesota	19	18	Arkansas	3	45
Georgia	19	18	Nevada	2	47
Michigan	17	20	North Dakota	2	47
Colorado	16	21	Utah	2	47
District of Columbia	16	21	Delaware	1	50
Tennessee	16	21	Vermont	1	50
West Virginia	16	21	Overseas American Schools	1	50
Missouri	16	21	Puerto Rico	0	53
Nebraska	15	26			
New Hampshire	12	27			
Washington	12	27			

Modified after a report from Science Service (6).

In comparing the data of Table 2, Figure 1 and Figure 2, it is clear that

1. total participation in the national Westinghouse Science Talent Search (WSTS) for senior students decreased by more than one-half between 1969 and 1971 (from 2361 to 1110);

2. between 1972 and 1982, participation in the WSTS has decreased slightly (from 1133 to 950);

3. participation in the Indiana Science Talent Search has declined between 1969 and 1982 (from 75 to 49), but since 1973, it has experienced a net increase, while participation in the WSTS has decreased.

There are many possible reasons for these declines. Perhaps the greatest reason for the drastic decline between 1969 and 1971 was the new regulation that vertebrate animal studies would not be considered in the Westinghouse Science Talent Search beginning in 1970-71. That regulation is still in effect today. In the Indiana STS between 1970 and 1982, 156 of 278 finalists (56.1%) submitted studies of a biological nature. Of these, at least 61 (39.1%) involved the use of vertebrate animals under the supervision of properly trained adults.

Other high achievers who study science and mathematics prior to grade twelve would benefit from conducting research and they should be encouraged to do this. The prospects of travel, recognition and financial aid associated with the WSTS, state searches, science fairs and other competitions can lead to broader particpiation in these activities. Successful participation can provide new purpose to the study of science and mathematics courses while in high school. They are also more

TABLE 2. *Comparison of the Indiana Science Talent Search and the Westinghouse Science Talent Search (1969-82)*

	INDIANA STS										WESTINGHOUSE STS				
					FINALISTS ONLY										
	NUMBER OF ENTRANTS	NUMBER OF FINALISTS	NUMBER OF WINNERS	JUNIOR FINALISTS	BIOLOGY TOPICS	CHEMISTRY TOPICS	PHYSICS TOPICS	ENGINEERING	MATH/COMPUTER	ALL OTHER TOPICS	TOTAL ENTRANTS FOR THE U.S.	INDIANA RESIDENTS	INDIANA FINALISTS (OF TOP 300)	PERCENTAGE OF FINALISTS FROM INDIANA	NATIONAL WINNERS FROM INDIANA (OF 40 IN U.S.)
1969	75	28		–	9	7	6	1	2	3	2361	86	7	2.33	1
1970	43	23	12	–	9	7	3	0	2	2	2075	56	3	1.00	0
*1971	38	18	10	–	7	5	2	0	2	2	1110	38	7	2.33	0
1972	17	12	9	–	6	0	2	2	1	1	1133	24	5	1.67	1
**1973	29	19	9	1	12	2	2	0	1	2	1105	25	5	1.61	1
1974	30	16	9	2	10	1	1	1	3	0	1104	24	7	2.33	0
1975	49	24	13	1	15	5	3	0	0	1	1118	30	7	2.33	0
1976	57	22	11	1	15	4	2	0	1	0	1013	26	9	3.00	1
1977	43	24	13	4	12	1	3	4	0	4	1009	23	8	2.67	2
1978	38	22	13	4	12	2	2	6	0	0	967	21	10	3.33	0
1979	34	23	13	4	11	3	3	2	0	4	980	20	10	3.33	0
1980	41	24	15	4	16	3	0	4	0	1	950	27	12	4.00	1
1981	46	25	14	4	15	3	1	3	0	3	993	22	7	2.33	0
1982	49	26	13	7	16	4	1	2	2	1	950	21	7	2.33	1
TOTALS (1969-1982)	589	306	154	32	165	47	31	25	14	24	16868	443 (2.6%)	104 (2.5%)	2.47	8 (1.4%)

*The First year that vertebrate animal studies were not permitted in Westinghouse STS.

**The First year in which eleventh grade students were permitted to enter the Indiana Science Talent Search. modified after a report by Science Service (6).

likely to pursue a career in science, mathematics or engineering as a result of such successes. Teachers who have not experienced the frustrations and the joys of working with students as they develop research projects are missing a great deal. The author would be happy to discuss this with teachers and students. Those who wish to learn of schools which have students successfully involved in scientific research year after year are invited to contact the author who will identify those schools and the key teachers involved there.

Directors of fifteen state science talent searches responded to a questionnaire prepared by this author in 1981. The results of that survey in terms of participation by young men and young women are given in Table 3. It is interesting to note that for the period of 1979-1981, the percentage of women participants in the Indiana Science Talent Search (44.8%) was greater than that of all other states which responded (34.3%) and greater than that of women's participation in the Westinghouse Science Talent Search (29.7%). In this connection, we in Indiana compare very favorably. But in terms of involving most young women in meaningful scientific research during

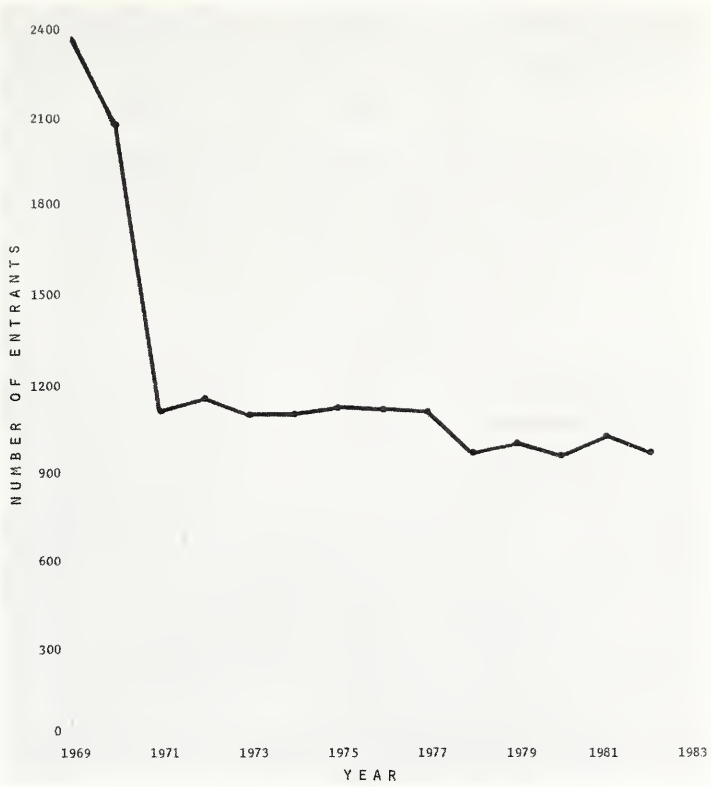
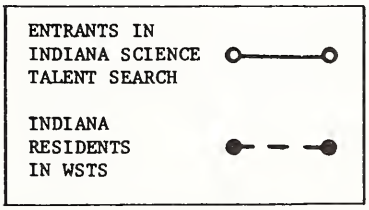


FIGURE 1. *Participation in the Westinghouse Science Talent Search Nationwide (1969-1982)*

the high school years, we in Indiana and the rest of the nation have much room for improvement.



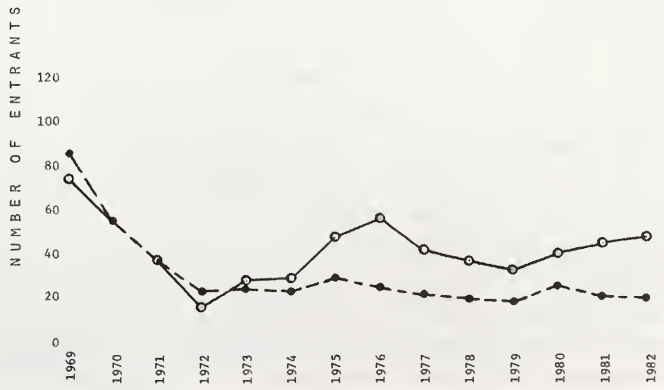


FIGURE 2. *Participation in the Indiana Science Talent Search and the Westinghouse Science Talent Search by Indiana Residents (1969-82)*

TABLE 3. *Westinghouse Science Talent Search Entrants by Sex*

State	1979-80 Women	1979-80 Men	1980-81 Women	1980-81 Men	For 2 Yrs (1979-81) Women	For 2 Yrs (1979-81) Men	% Women
Alabama	16	24	17	20	33	44	42.9
Colorado/Wyoming	6	8	5	6	11	14	44.4
Florida	16	43	9	24	25	67	27.2
Indiana	14	27	25	21	39	48	44.8
Iowa	2	1	4	14	6	15	28.6
Kansas	3	6	2	3	5	9	35.7
Maine	Not	Known	—	—	—	—	—
Mississippi	1	3	2	6	3	9	25.0
Montana	2	3	2	3	4	6	40.00
Nebraska	Not	Known	—	—	—	—	—
North Dakota	0	1	0	2	0	3	0.0
Utah	1	4	2	2	3	6	33.3
Virginia	Not	Known	—	—	—	—	—
Washington	Not	Known	—	—	—	—	—
West Virginia	Not	Known	—	—	—	—	—
Totals (Entrants in 15 state searches)					129	221	36.9%

All Entrants in WSTS (Not Compiled; Estimated to be About 33% Women)

	Women	Men	%Women
Honors Group, WSTS (1978-79)	80	220	26.7%
Honors Group, WSTS (1979-80)	100	200	33.3%
Honors Group, WSTS (1980-81)	86	214	28.7%
Honors Group, WSTS (1981-82)	90	210	30.0%
Totals (WSTS) 4 Years	356	844	29.7%

Based upon a survey of selected state science talent searches (1).

SCIENCE AND ENGINEERING FAIRS—REGIONAL (Usually grades 7-12, but some regional fairs accept elementary school students). These competitions are open to any public or parochial student within a designated region of a state. Usually, two winners from each regional fair are selected to attend the International Science and Engineering Fair. Other awards include ribbons or medals, books, cash prizes, hand calculators and recognition certificates. The student must display an exhibit of his or her investigation. For information, see your science teacher or guidance counselor.

SCIENCE FAIRS—LOCAL, DISTRICT, SCHOOL (Grade levels vary). These are competitive exhibitions open to students within a particular district, county, township or school for the purpose of selecting regional science fair entrants and to award local prizes. For information, see your science teacher or guidance counselor.

SCIENCE AND ENGINEERING FAIR—INTERNATIONAL. This is a competitive exhibition open to any public or parochial student in grades 9-12 who has been selected through regional fair competition (two from each region). Two senior division winners from each of about 200 regional fairs with their teacher sponsors and many fair directors will spend 5 to 6 days in the host city with all of their expenses paid. Student mixers, tours of scientific facilities and award banquets are provided. After the students' research investigation and exhibits are evaluated, winners receive scholarships, opportunities to work in government research laboratories, cash awards, trips and nationwide recognition. For information, write:

Director, International Science & Engineering Fair
Science Service
1719 N Street, N.W.
Washington, D.C. 20036

A list of possible science projects is available at $1.00 each (5), and a very good booklet which includes guidelines for student research is available from Moore Publishing Company (4).

SCIENCE STUDENT TRAINING PROGRAMS. Summer science training programs at various colleges and universities are open to those students who have completed the 10th or 11th grade. These programs last from one to ten weeks. Most offer in-depth instruction in one or more subjects such as astronomy, biology, chemistry, mathematics or physics. Many allow participants to participate in scientific research; some include college credit. While some are for commuters only, many involve residence on the college or university campus which adds another dimension to this experience. In most cases, funds are available to assist students whose families cannot afford the total cost of the program.

Today, seven students from the 1982 High School Science Student Institute at Indiana University are presenting the results of their research in four divisions of the Senior Academy, and at least four other participants in this Institute are presenting papers today in the Junior Academy of Science.

Information may be obtained by requesting the "Directory of Science Training Programs for High Ability Senior and Junior High School Students" (7) from:

Science Service
1719 N Street, N.W.
Washington, D.C. 20036

The new directory should be available in January. The deadline for return of applications to the project director is usually March 15 or April 1. Students should apply early.

JUNIOR ACADEMY OF SCIENCE. This is an affiliate organization of the Academy of Science of most states, open to science clubs at the junior or senior high school level. Students interested in student-research are encouraged to form a research group or science club and ask their sponsor to apply for club membership in their state. Primary emphasis is upon presentation of research papers for peer evaluation with opportunity for discussion with members of the senior Academy of Science as well.

In Indiana, contact: Leota Skirvin Smith, Director
Indiana Junior Academy of Science
State Road 46 S
Nashville, Indiana 47448

In other states, contact:
American Association for the Advancement of Science
Association of the Academies of Science
1515 Massachusetts Avenue, N.W.
Washington, D.C. 20005

JUNIOR ENGINEERING AND TECHNICAL SOCIETY (JETS). This competition involves *teams* of students in grades 10-12 in biology, chemistry, English, graphics, mathematics and physics. Team trophies and individual awards are presented to the outstanding performers. In Indiana, contact:

Lester J. Harris
State Coordinator, JETS
2918 East Eighth Street
Anderson, Indiana 46012

NATIONAL JUNIOR SCIENCE AND HUMANITIES SYMPOSIUM. Students in Indiana and Eastern Illinois who have been nominated by their schools will be invited to attend and describe their research during a three-day meeting to be held at Indiana State University in Terre Haute, Indiana, each April. From this group, finalists will be selected to present papers at the national symposium which is usually scheduled for May. For more information, write:

Dr. Kenneth Uhlhorn
Science Teaching Center
Indiana State University
Terre Haute, Indiana 47809

Students outside Indiana may contact Barbara Osbourn (see address below).

INTERNATIONAL YOUTH SCIENCE FORTNIGHT. This program consists of a two-week conference held each summer in London, England, involving approximately 450 students from 20 to 25 countries who have been selected from various national junior science and humanities symposia. The aim of the fortnight is to provide a deeper insight into science and its applications for the benefit of mankind and to develop greater understanding between young people of all nations. The program includes lectures by eminent scientists, demonstrations, visits to research laboratories and industry, panel discussions and social activities. For information, write:

Barbara Osbourn, National Coordinator
Junior Science & Humanities Symposium
Duke University, East Campus
Durham, North Carolina 27708

SPACE SHUTTLE STUDENT INVOLVEMENT COMPETITION. The purpose of the Space Shuttle Student Involvement Project, sponsored by the National Aeronautics and Space Administration (NASA) and the National Science Teachers Association (NSTA), is to stimulate interest in the science and technology of space by students in grades 9 to 12 in the U.S. and its territories. Up to 20 regional winners will be selected in each of ten regions; from these, up to 20 students will be selected for national honors. Regional winners and their teacher-sponsors will be invited to participate in a conference in the spring at one of the NASA research facilities with expenses paid. Each entry should be a typed proposal of not more than 1000 words describing an experiment or demonstration which could be performed by astronauts in the course of a space shuttle mission. The proposals should be received by the regional director by February 1. Entry

materials, rules booklets and a listing of the regional directors may be requested by teachers from:

Space Shuttle Student Project
National Science Teachers Association
1742 Connecticut Avenue, N.W.
Washington, D.C. 20009

INDIANA ACADEMY OF SCIENCE SPEAKERS' BUREAU. The Indiana Academy of Science provides a list of scientists and engineers who are willing to speak on scientific and engineering matters to schools, clubs and other community organizations. For information, write:

Walter Cory, Director of Public Relations
Indiana Academy of Science
Education Building 253
Bloomington, Indiana 47405

FUNDS FOR HIGH SCHOOL STUDENT RESEARCH. In addition to these examples, you might contact other organizations for possible support.

1. The American Heart Association, Indiana Affiliate, annually provides twenty-three $100.00 cash awards and certificates to high school students for research under the T. A. Kleckner Science Grant Awards Program. The research investigation need not be related to the heart or circulatory system. Awards are provided through a state-wide competition open to any high school student. Applications must be in the offices of the Indiana Heart Association no later than October 29. Winners are announced by about November 15. For further information write:

David Livengood
American Heart Association, Indiana Affiliate
222 South Downey Avenue
Indianapolis, Indiana 46219

2. The American Lung Association of Indiana Research Grants and Scholarships offers students in grades 10-12 an opportunity to compete for eight research grants of $150.00 each, one scholarship of $1,5000.00 and one of $500.00. The school sponsoring the top scholarship award recipient will also receive an award. The program, which began in 1974, has been very successful. Proposals to support pulmonary-circulatory research are due October 22. Detailed information may be obtained from:

Managing Director
American Lung Association of Indiana
30 East Georgia Street, Room 401
Indianapolis, Indiana 46204

3. The research grants committee of the Indiana Academy of Science provides funds for research investigations by secondary students. Science club activities of an investigative nature will be considered as well as those of individual members. A brief description (200-500 words) of the project's budget and objectives is required. Requests, ranging from a minimum of $50 to a maximum of

$250, must be made by a sponsoring teacher of the club and be signed by the principal or superintendent of the school system. Proposals from junior schools and senior high schools in Indiana only should be submitted before June 15. Science clubs and individuals who receive activity grants are required to submit a report of their activities to the Youth Activities Committee Chairperson by May 1 of the following year. Address all inquiries and proposals to:

Dr. Ernest Campaigne, Chairman
IAS Research Grants Committee
Department of Chemistry
Indiana University
Bloomington, Indiana 47405

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Note: The efforts of previous coordinators for school science programs at Indiana University and others who contributed to the programs described here are gratefully acknowledged.

Research, Writing, and the Computer in Introductory Biology

Gary E. Dolph
Division of Liberal Studies
Indiana University at Kokomo, Kokomo, Indiana 46902

The most difficult task facing an introductory biology teacher is to teach creative thinking and hypothesis testing. This task is most often relegated to the laboratory, where students carry out laboratory exercises and write laboratory reports. These activities, however, rarely teach how a scientist works. Laboratory exercises are useful for teaching techniques, but they are not flexible enough to encourage hypothesis testing. Because laboratory reports are artificially structured for ease of grading, they do not allow a student to see what technical writing is really like.

By combining the computer, the library, and the laboratory, the biology instructor can assign research projects requiring creative thinking and hypothesis testing. This approach is currently in use in Introductory Biology at Indiana University at Kokomo. The computer can provide data from experiments which are too expensive, too dangerous, or too time-consuming to be carried out in the laboratory. For example, an experiment dealing with total plant energy budget and photosynthesis would be too expensive; one dealing with disease-causing pathogens would be too dangerous; and one dealing with a multiple site ecological study would be too time-consuming. The data required by these experiments could be provided quite easily using the computer. The library can provide the background information required by the students to understand the experiment. Pertinent references could be placed on reserve or the students could be taught to carry out a literature search using biological source materials. The techniques required to collect the data can be illustrated by actual laboratory exercises or by slides or films to give the students some idea of what type of work the computer program is doing for them. Finally, the science report prepared at the end of the assignment can be used to teach the students how an actual paper is prepared without the artificiality of being a laboratory report.

A computer-based research project provides a very valuable learning experience for both majors and nonmajors. Their experience with this project is the most important concept they can take away from an introductory course. After working on an actual research project, the nonmajors should feel more secure about the accuracy and objectivity of scientific reports. Majors can use this experience to decide whether or not they would like to perform this type of work for the rest of their lives.

The Model

I chose an ecological model as the basis of the research paper. Plant ecologists (9) and paleobotanists (2,3,11) have postulated that a close relationship exists between leaf form (either leaf margin type or leaf size) and climate (mean annual temperature or average annual precipitation). Bailey and Sinnott (2,3) showed that the percentage of woody dicot species having leaves with entire margins (neither toothed or lobed) decreased with decreasing mean annual temperature either altitudinally or latitudinally. Their conclusions have been supported by more recent work by Wolfe (11). The relationship between leaf size and climate is less clear. Raunkiaer (9) postulated that leaf size in the woody dicots should

decrease with decreasing average annual precipitation either altitudinally or latitudinally. In contrast, Givnish (7) predicts that average leaf size should be highest in the tropics, decrease into the subtropics, increase once more into the temperate zone, and finally decrease toward the poles. These theories provide an excellent basis for a research project. Sufficient disagreement exists between workers in the field such that no certain answers are available in the literature. Each theory deals in a general way with the variation of leaf form with climate. Only one detailed study has been carried out over a small area (6). Therefore, a locally relevant project could be developed, if the students studied the climate and flora of their State.

Three simulations are available enabling my students to study the variation in leaf form and climate in Indiana: CLIMATE, LFMARG (LeaF MARGin), and LFSIZE (LeaF SIZE). CLIMATE provides weather data from 91 weather stations in Indiana and the surrounding states of Michigan, Ohio, Kentucky, and Illinois. Seven climatic parameters may be studied: mean annual temperature, mean annual range in temperature, average annual precipitation, mean annual biotemperature (8), potential evapotranspiration ratio (8), effective temperature (1), and equability (1). Climatic data may be requested for a specific series of months (for example, May to October, the growing season in most of Indiana) or for the whole year. LFMARG calculates the percentage of species having leaves with entire margins for any county in Indiana. LFSIZE calculates either the average leaf area (7) or a seven-class leaf size distribution (9) for any county in Indiana. LFMARG and LFSIZE calculate answers using either the total number of woody dicotyledonous species found in a county or some subset of this information. An analysis of the total woody dicotyledonous vegetation may be subdivided into trees, shrubs, or vines based on growth form or into deciduous or evergreen species based on plant habit. Therefore, a student might study the variation of only the evergreen trees of Indiana.

These three simulations are built around two large data bases. CLIMATE queries a data base built using climatic data provided by the U.S. Department of Commerce (10). Their report gives the average temperature (°F) and the average precipitation (inches) by weather station for each month of the year. After being converted to the metric system, these data may be used directly, or they may be used to calculate a climatic index, such as mean annual biotemperature. LFMARG and LFSIZE query a data base containing information on each of the 227 woody dicots native to Indiana. The data bank records the name of each species, its leaf characteristics (both margin type as well as the range in leaf length and width), its growth form (tree, shrub, or vine), its growth habit (deciduous or evergreen), and the counties in Indiana in which it grows.

The Assignment

The students have approximately ten weeks to complete their research projects. Two weeks after the start of the semester, the students are shown the University's computer facilities and are introduced to CLIMATE. Initially, the students use CLIMATE to obtain information to map the variation in mean annual temperature and average annual precipitation over Indiana. They may not collect data from all 91 weather stations. They may request information for a total of 30 weather stations, but they are asked to collect the data slowly while reflecting on where they should sample in order to gain an accurate picture of how the climatic variable being studied fluctuates across Indiana. After they start analyzing the variation in leaf form, they may return to CLIMATE to study other

climatic variables or to study the effect of temperature and precipitation over a portion of the year. At the end of two weeks, the students must turn in their contour maps for the first two environmental variables. The maps are not graded, but the students are told where corrections should be made.

At the same time as the students are starting their computer projects, the basic principles of ecology are being covered in lecture. In addition, the students spend two weeks identifying the woody plants on campus in the laboratory. After learning how to identify these plants, they analyze a forest stand to identify the most important species. At the end of the first five weeks of class, the students have lecture, laboratory, and computer experience with their projects.

Once their climatic maps have been completed, the students are ready to start studying the variation in leaf form in Indiana. However, except for some background material that has been presented in lecture, they do not know what to expect. To obtain background information, the students must go to the library and answer a number of questions which I provide. For example, a student studying the variation in leaf margin must answer the following questions:

1. Who were the first researchers to study the relationship between leaf margin type and environment?
2. What did the first researchers conclude?
3. What other researchers have studied this relationship?
4. Some researchers feel there is a close relationship between leaf margin type and environment. How close?
5. Does anyone oppose these views? Who? What does he/she believe?
6. You will be studying the variation in leaf margin type and climate at the local level in Indiana. In what other states has this relationship been studied? What conclusions were reached relative to the subset of the vegetation of Indiana you might be studying?

The articles needed to answer these questions (2-7,9,11) are on reserve in the library. By answering these questions, the students collect the information necessary to write the Introduction of their research report. Two weeks after the readings are assigned, the Introduction must be turned in. The Introductions are then passed out in class, and the students criticize their own writing.

The students use their Introductions to reserve the type of leaf variation they would like to study. One student might choose to study the variation in average leaf size for the total woody dicotyledonous flora of Indiana. Another student might wish to analyze the leaf margin variation in only the deciduous shrubs. After choosing their projects, the students are introduced to the simulations, LFMARG and LFSIZE, which will provide them with information about their projects. The students will analyze the vegetation in only 30 out of the 92 counties in Indiana. Only a portion of the counties in Indiana may be studied because complete sampling is impossible in an actual field study due to time and financial constraints. If a student is particularly insistent, he/she may analyze more than 30 counties. This student is warned that simply adding more data does not necessarily make it easier to arrive at valid conclusions. After finishing their analyses, the students, on a map of Indiana, contour the variation encountered. As with the climatic maps, the vegetation maps must be turned in for approval.

The students reach conclusions by comparing the variation on their climatic maps with that on their vegetation maps and noting any trends. An interesting feature of the simulation is that the general theories relating leaf form with climate are not necessarily accurate at the local level in Indiana. For example, leaf margin variation for the dicotyledonous tree species conforms fairly well to the variation

in mean annual temperature, but leaf margin variation in the woody shrub species does not. Because the results they find do not always fit the theories in the literature, the students must give careful consideration to the conclusions they draw in their final reports.

The final report must be written in the format of a scientific journal. The report must contain the following sections and answer the following questions:

1. Title	What was the experiment about?
2. Introduction	What work has been done on this topic? How is your project related to this work?
3. Materials and Methods	How was the project carried out?
4. Results	What did you discover?
5. Conclusions	Do your results confirm or refute what other researchers have said?
6. Literature Cited	What references did you use?

Only after the final report has been turned in is the student's project graded. Two points need to be mentioned about how the project is graded. First, although the conclusions are important, their grade is not based upon obtaining the "correct" answer. The lack of emphasis on a "correct" conclusion is a reflection both of the complexity of the project and of a student's ability to argue in favor of some plausible conclusion. Second, considerable emphasis is placed on how well the report is written, reflecting the need to have science reports graded for grammar and style.

Conclusions

Nonmajors as well as majors taking a course in science should come away from that course with some understanding of how a scientist in that discipline functions. Neither the lecture nor the laboratory provides this experience. The computer-based research project described in this paper does. By combining the lecture, the laboratory, the library, and the computer, this project illustrates the steps in the scientific method and how an individual scientist struggles to apply them to his or her research. In addition, the project does not neglect technical writing, the method by which the information generated reaches the scientific community.

Acknowledgment

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A Survey of Introductory Biology Laboratory Manuals for Content and Experimental Inquiry

THOMAS A. FOGLE and ERIC D. MOULD
Department of Biology, Saint Mary's College, Notre Dame, Indiana 46556

Introduction

A laboratory experience in the sciences can serve to: teach or reinforce concepts, learn techniques, develop inquiry skills, and convey how science is carried out. The techniques and concepts employed in the lab will depend on the subject matter, whereas inquiry and general scientific understanding do not. The latter two educational goals are interrelated; inquiry is an integral part of the methodology of science. Recent investigations on pre-college science curricula (6) and secondary laboratory handbooks in science (5) have detailed the role of inquiry. Essays by Mills (2) and Postlethwait (4) have focused attention on this goal in college biology courses.

Reports on the subject matter of introductory biology laboratory exercises and the extent to which scientific experimentation is used as a vehicle to convey scientific inquiry are sparse. As a means of assessing the direction taken in laboratory education for college biology courses, we have conducted a survey of published laboratory manuals to 1) identify the major topics emphasized and 2) ascertain the degree to which experimentation is utilized.

Sample

Twenty published laboratory manuals, organized to complement an independent course in introductory biology, were chosen for review (see Appendix). In order to make meaningful comparisons, manuals designed expressly for non-majors were not considered. The manuals sampled have all been published between 1977 and 1982 and represent authorship from three junior colleges, five colleges and, twelve universities. It is reasonable to assume that by virture of being published, the exercises have been well thought out, tested in the laboratory, and are actively used. No conscious effort was made to select manuals based on content or style. Although the manuals were evalutated individually, the data were analyzed collectively and judgements were not made on any one manual.

Method of Survey

The manuals designed for one-term and two-term courses in biology were analyzed separately for content, but not for inquiry. Since the organization of manuals varies widely, no one system of subdividing the discipline would satisfy all manuals. Therefore, a hierarchial division of biology was chosen for convenience, and any given laboratory unit could be listed in more than one topic. This scheme allows generalizations to be made about content without detailing endless minor differences between manuals.

Six major areas, twenty-one topics, and thirty-six subtopics were identified. Many additional subtopics could be listed, but this set has the distinction of being easily definable and readily comparable from manual to manual. No attempt was made to objectively measure depth of coverage within topics or subtopics.

The level of scientific inquiry, as reflected by experimental analysis, was classified into one of three categories:

1. conceptual or informational experience;
2. data manipulation and interpretation exercise; and
3. student designed experimental analysis.

Category one situations consist of well outlined procedures for students to follow and would include, among other things, demonstrations, dissections, and observations of cell types or representative organisms from different phylogenetic groups. Category two is characterized by student involvement in a planned experiment which must be interpreted utilizing gathered data. Category three is defined as a student formulated experimental design that is carried out to its conclusion. This scheme is not intended to measure components of higher order thought, such as the need to conceptualize, synthesize, or use abstract reasoning. Rather, the focus is on the application of the scientific method by the student. This subsumes higher order thinking to a lesser degree in level two and to a higher degree in level three. Level one may or may not entail higher levels of thought, but is distinguished by the fact that the student is not expected to conduct an experiment and gather data.

Results

Table 1 provides a breakdown of the discipline into six major areas within which topics and subtopics are listed. In general, topics occur more frequently in two-term manuals with the exception of those in the cellular/molecular biology area but, both sets of manuals show a similar hierarchial type of organization.

Heavy emphasis is given to proper use of the microscope in most manuals with greater detail provided in most two-term manuals. No other instrumenta-

TABLE 1. *Coverage of topics in published lab manuals designed for one or two-terms of majors biology*

Major Area / Topic / Subtopic	Percent of Manuals	
	One-Term	Two-Term
Cellular/Molecular Biology		
Use of Microscope	83	79
Magnification/ Measurement	50	64
Slide Preparation	50	100
Dissecting Scope	0	43
Oil Immersion	0	14
Biochemical Analysis	100	57
Carbohydrates	100	57
Lipids	67	50
Protein/Amino Acids	100	50
Nucleic Acid	0	21
Enzyme Analysis	67	71
Simple Kinetics	33	57
Parameters Affecting Activity	50	71
Cell Biology	100	100
Plant/Animal Structure	100	100
Diffusion	67	100
Osmosis	83	100
Active Transport	17	29
Organismal Biology		
Metabolism	67	93
Aerobic Respiration	17	79
Anaerobic Respiration	67	57

TABLE 1.—Continued

Major Area / Topic / Subtopic	Percent of Manuals	
	One-Term	Two-Term
Vertebrate Anatomy	67	100
Digestive System	67	86
Respiratory System	17	71
Nervous System	17	71
Circulatory System	50	100
Skeletal System	17	64
Muscular System	17	43
Excretory/Reproductive	50	79
Vertebrate Physiology	50	79
Human Senses	50	64
Stimulation	0	36
Behavior	33	43
Taxis	17	36
Vertebrate Observation	17	36
Invertebrate Observation	17	29
Embryology/Development	67	93
Cell Division/Genetics		
Mitosis/Meiosis	83	100
Genetics	100	93
Human Genetics	67	79
Mendelian Problems	100	79
Drosophila	17	57
Population	33	29
Botany		
Plant Structure	100	100
Photosynthesis	100	100
Isolate Pigments	100	86
Physiology	17	71
Growth	67	57
Diversity		
Animal Survey	33	100
Plant/Fungi Survey	50	100
Bacteriology	50	86
Use of Dichotomous Key	17	29
Ecology/Population Biology/Evolution		
Ecology	50	79
Outdoor Experience	17	50
Monitor Population	17	50
Ecosystem Modeling	17	14
Evolution	17	21

tion is so consistently designated for special explanation. Biochemical testing is not as commonly incorporated in two-term manuals as compared to one-term manuals, but this difference maybe is due to the greater liklihood that such methods would be included within the context of some other topic. For example, a glucose determination or analysis of starch breakdown might be found in an exercise on digestion. No doubt the bias toward certain biomolecules, is based on a conscious choice that certain methods are more amenable to laboratory exercises than others. The central role that enzymes play in coordinating rate of reactions, and therefore living processes, is reflected in the pervasive application of enzyme analyses in the two sets of manuals.

Universal inclusion of cellular observation is not surprising since cells are fundamental units of life, wet mounts are easy to make, and prepared slide collections are available in most institutions. Similarly, diffusion and osmosis con-

cepts are deemed important, while being simple and cost-effective to perform in the laboratory. However, experiments on active transport are much less commonly included. Usually the experiment suggested is the effect of congo red dye on live and dead yeast cells.

Aerobic metabolism experiments can be tested with plants (two manuals), animals (two manuals), or both (seven manuals), while experiments in anaerobic metabolism typically utilize yeast. Experiments from one-term texts tended to do either an anaerobic or aerobic exercise, but not both.

The frog and fetal pig are used with equal frequency for vertebrate dissection. Among the biological systems, circulation and musculature are the most and least commonly dissected, respectively. Selection of organ systems can be influenced by the choice of organism. The respiratory, excretory, and reproductive systems are more often analyzed if the fetal pig is used, whereas the skeletal, muscular and nervous systems are more typical of frog dissections. The investigation of the digestive and circulatory systems was apparently not biased by the choice of specimens.

With the exception of aerobic metabolism exercises, vertebrate physiology exercises receive less emphasis than vertebrate anatomy. Physiological processes, when included, often entailed a study of human senses. More sophisticated experiments, such as electrical stimulation of muscle tissue, are the rare exception. Here again, there is a tendency to use simple, inexpensive exercises which necessitate a modicum of equipment.

Embryology is incorporated into all but two manuals. Inclusion of more than one type of organism is common; three manuals use three organisms each and eight use two, yet only two manuals have the same combination of specimens. The number of manuals using each type of specimen are; chick 10, frog 10, starfish 7, and sea urchin 4.

Both genetics and mitosis-meiosis labs are highly represented in our sample. Most commonly, genetics labs demonstrate human genetics traits and, to a lesser degree, utilize *Drosophila* exercises. Most exercises are supplemented with a heavy dose of Mendelian problems.

Plant structure and photosynthesis are found in every manual sampled. However, physiological processes other than photosynthesis do not receive the attention afforded structure.

Phylogenetic surveys of either plants or animals are extremely diverse in depth. On the one hand, one-term manuals may demonstrate one or two organisms from each phyla, on the other hand two-term manuals often devote one whole unit to a single phylum. Of the two-term manuals, 12 of 14 recommend invertebrate dissections. Among this group, dissections of the earthworm, crayfish, clam, starfish and grasshopper are each suggested in 67% or more of the manuals. Other dissections, such as *Ascaris* (30%) and the squid (9%), were also recommended as well as a diverse group of chordate dissections (other than the frog and fetal pig) including the bird, shark, eel, rat, and *Amphioxus*. Plant diversity and plant reproduction are generally intertwined so that a unit bearing either title often serves as the vehicle to teach the other. For the purposes of this survey, both are listed under plant diversity.

Table 2 compares biological topics with respect to the level of independence required by students when performing experiments. The percentages listed are computed from the set of manuals that included each topic. The topics are placed into three groupings according to the combined percentage of categories two and three, and ranked from highest to lowest. It is evident that data gathering and interpretation permeate most lab topics, but that student originated experimen-

TABLE 2. *Biological topics categorized for the level of student involvement in experimentation.*

	Percent of Lab Manuals in each Category		
Topic	1	2	3
Enzyme Analyses	13	80	7
Ecology	23	62	15
Plant Physiology/Growth	29	57	14
Metabolism	32	68	0
Behavior	38	50	12
Genetics	42	53	5
Biochemical Analyses	47	53	0
Photosynthesis	50	45	5
Cell Properties	55	40	5
Mitosis/Meiosis	85	15	0
Plant Structure	90	10	0
Vertebrate Physiology	94	6	0
Plant/Animal Surveys	100	0	0
Embryology/Development	100	0	0
Anatomy	100	0	0
Cell Structure	100	0	0
Evolution	100	0	0

tation is rare. Indeed, most of the category three experiments were clustered in three of the twenty manuals. Ford *et al.* (10) lists "challenges" at the end of many units, some of which fit category three; but, since these are relegated to optional status, they were not tabulated.

Discussion

When one-term and two-term manuals are compared, both place a high priority on broad coverage rather than emphasizing depth in a limited range of topics. This is supported by the fact that time constraints are much more severe for a course using a one-term manual, yet the reduction in the percent of lab manuals teaching most topics is small. Despite the fact that depth and organization vary considerably, 90% of the manuals included exercises on cell structure, use of the microscope, and biomolecules among two of the first three units and 60% concluded with exercises on ecology or population biology. This suggests a hierarchial scheme, although the sequence of topics between the beginning and the end is so variable as to preclude any trend. Except for the light microscope (and to a lesser degree a spectrophotometer), lab equipment necessary to complete most all exercises is minimal. Perhaps this is related to the predominance of exercises on structure (e.g. dissections) over those involving processes (e.g., organismal physiology).

The inquiry method of learning has been touted by educators for years (1). Welch et al. (6) defines inquiry as a way of thought in which "human beings seek information or understanding." They recognized scientific inquiry as a subset of general inquiry and designated three themes within this subset, one of which, "science process skills," comes closest to the categories two and three we have outlined. Their study of pre-college science education reaffirmed their view that scientific inquiry was needed in the classroom, but that the current state of affairs was less than desirable. An evaluation of secondary school laboratory handbooks using a scaling system comparable in goals to our own (5), resulted

in strikingly similar findings. The findings reported that students are often expected to gather data and interpret results, but seldom are asked to "work according to their own design."

There is concern for scientific inquiry in college science courses as well, though the methods to tackle the problem are varied. Molls and Allen (3) have found that students in introductory biology display improvement in critical thinking ability and content knowledge when confronted with data to interpret. The students use short video tape presentations followed by planned, but non-directive discussions. Postlethwait (4) has opted for a multifaceted approach which includes, among other things, an extended research project, oral presentations, readings in journal articles, and written reports. Mills (2) reports his laboratory experiences in introductory biology in which he integrates student originated experiments throughout a two semester course. He argues persuasively that this method is highly motivating to students without limiting breadth, being expensive, or causing a severe drain on the instructor's time. He finds that 20% of laboratory time can feasible be spent on independent investigations. Although Table 2 cannot be translated into a distribution of laboratory time, our subjective opinion is that all manuals in the survey fall well below this level.

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6. WELCH, W. W., KLOPFER, L. E., AIKENHEAD, G. S. AND J. T. ROBINSON. 1981. The role of inquiry in science education: Analysis and recommendations. Science Education 65:33-50.

Appendix

One-term Manuals

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3. LATTA, V. G., MCDONALD, C. A., AND J. W. ROBERTS. 1977. Principles of Biology: Laboratory Investigations. Hunter Publishing Co., Winston-Salem 138 pp.
4. SALTER, C. G. 1979. Biology Laboratory Manual. Hunter Publishing Co., Winston-Salem, 142 pp.
5. UBENHAUN, L. M., NORD, R. P., WEEKS, T. F., AND R. W. TYSER. 1980. Principles of Biology Laboratory Manual. Burgess Publishing Co., Minneapolis, 222 pp.
6. WYNN, T. E. 1979. Biology in the Modern World, 2nd ed., Hunter Publishing Co., Winston-Salem, 178 pp.

Two-term manuals

7. EBERHARD, C. 1982. Biology Laboratory - A manual to accompany Biology, 2nd

ed., Saunders College Publishing, New York, 328 pp.
8. ENGER, E. D., GIBSON, A. H., KORMELINK, J. R., ROSS, F. C., SMITH, R. J. BORGMAN, C. H., AND R. H. NORTHRUP. 1982. Concepts in Biology, 3rd ed., Wm. C. Brown Company Publishers, Dubuque, 162 pp.
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15. SANDERS, G. P. 1981. Laboratory Manual, Biology: The Science of Life. Scott, Foresman, and Company, Glenview, 372 pp.
16. STETLER, D. A. AND J. W. VIA. 1977. Biology Laboratory. Hunter Publishing Company, Winston-Salem, 218 pp.
17. URBAN, E. K., BICKERT, J. H., BAROUTSIS, J. G., STULLKEN, R. E., and J. B. BLACK. 1978. Laboratory Manual for Introductory Biology. Hunter Publishing Company, Winston-Salem, 167 pp.
18. WEISZ, P. B. 1982. Laboratory Manual in the Science of Biology, 5th ed., McGraw-Hill Book Company, New York, 279 pp.
19. WINCHESTER, A. M. 1979. Biology Laboratory Manual, 6th ed. Wm. C. Brown Co., Publishers, Dubuque, 331 pp.
20. WODSEDALEK, J. E., DEAN, H. L., AND T. E. ROGERS. 1980. General Biology Laboratory Guide, 4th, ed., Wm. C. Brown Company Publishers, Dubuque, 266 pp.

A Survey of Laboratory Manuals in Physiology

Eric D. Mould and Thomas A. Fogle
Department of Biology
Saint Mary's College, Notre Dame, Indiana 46556

Introduction

Laboratory sessions are extremely valuable assets in the teaching of physiology because they elevate the students from passive absorbers of information to active participants in their own learning. "Hands-on" manipulation of instruments and biological material also reinforces basic concepts and develops expertise in physiological methodologies. Successful laboratory experiences can arouse student interest in subject matter developing excitement in the study of life and, ultimately, foster conceptual and manipulative skills.

Potentially, there exist literally hundreds of laboratory experiences which could be conducted on each organ system. However, our academic experiences have indicated that the laboratory offerings in physiology courses are relatively consistent. Temporal, financial, and instrumental limitations can play important roles in limiting the scope of exercises utilized by teaching laboratories in college level physiology. Additionally, tradition dictates that laboratory exercises "work" and are within the expertise of the students' instrumental, animal handling, and surgical skills. The purpose of this survey is to document the breadth and extent of occurrence of subjects and methodologies in published physiology laboratory manuals for college courses.

Methods

Nine human physiology and eleven general physiology laboratory manuals were reviewed for this study (Appendix). Grouping of the manuals into these headings was directly related to the purpose of the manuals as stated by the authors in the "Forward" sections of the texts. Without exception, human physiology manuals were designed for one term courses for preclinical or physical education students while general physiology manuals were intended for upperclassmen majoring in life sciences. Selection of manuals was based on their inclusion in the latest edition of *Books in Print* and availability from the publishers. Although this selection procedure cannot be considered either random or complete, we do believe they are recent and representative of the laboratory schemes utilized by many physiology laboratories.

All manuals were read in their entirety and each exercise was catagorized into one of the major topic and subtopic headings for Human (Table 1) and General (Table 2) Physiology. We developed topic areas which were based primarily upon organ systems. These criteria for topic breakdown proved to be judicious since laboratory units as they appeared in the manuals rarely involved more than one organ system. Subtopics were developed on the basis of which methodologies were utilized under each topic heading. Since there existed considerable variety in the depth of coverage in the topic areas, the numbers of subtopics were not consistent within or among manual types.

Laboratory exercises were also assigned to one of three categories of "independent student involvement." The categories were:

1—Students were carefully directed by the manual in both data gathering and interpretation.

TABLE 1. *Percentage of Human Physiology laboratory manuals including exercises in each topic and subtopic.*

Topic / Subtopic	% Manuals With: Topic	Subtopic
Hematology	100	
Histology		100
Hematocrit		100
Blood Type		100
Clot Time		86
Hb		57
Blood pH		29
Sedimentation		29
Electrophoresis		14
Circulation	100	
Blood Pressure		100
Heart Sounds		86
EKG		72
In Vitro		43
Microcirculation		43
Ergometry		14
Neurophysiology	100	
Reflexes		100
Vision		100
Audition		100
Taste		71
Olfaction		43
Cutanious		14
EEG		14
Renal Physiology	100	
Urinalysis		100
Homeostasis		86
Renal Clearance		14
Respiration	100	
Spirometry		86
Pnemography		57
Effect-CO_2		43
Sounds		43
Ciliary Movement		29
Calorimetry		29
Cellular Processes	100	
Osmosis		100
Enzymes		29
Cell Respiration		29
Karyotype		14
Active Transport		14
pH Regulation		14
Nutrition	86	
In Vitro Digestion		86
Smooth Muscle		43
GIT Sounds		14
Muscle Physiology	86	
Contraction		86
EMG		14
Endocrinology	57	
Epinephrine		29
Insulin		29
Thyroid Hormone		14
Sex Steroids		14
Reproduction	43	
Antigen Test		14
Bioassay Test		14
Fertilization		14

2—Students were required to manipulate and interpret data.

3—Students developed at least portions of the experimental design.

We quickly found that one must be extremely careful not to associate a hierarchy of thought processes with this scheme. Many areas covered by the manuals can only be effectively covered by the category 1 mode (e.g., histology of blood cells) while some areas readily invite the student to participate in experimental design (e.g., effects of various parameters on ventilation in rats). However, many experiments can be molded to neatly fit any of the three categories at the discretion of the authors. For example, a laboratory on peripheral blood pressure could involve a description of technique and measurement of blood pressures (category 1), measurement, comparison and interpretation of blood pressures taken under variable conditions (category 2), or the student could be required to design an experiment to demonstrate what factors effect peripheral blood pressure (category 3).

TABLE 2. *Percentage of General Physiology laboratory manuals including exercises in each topic and subtopic.*

Topic / Subtopic	% Manuals With: Topic	Subtopic
Circulation	100	
Cardiac Cycle		100
EKG		100
Blood Pressure		89
Exercise		56
Microcirculation		56
Heart Sounds		44
Direct Blood Pressure		33
Pleysmography		33
Blood Volume		11
Comparative Systems		11
Neurophysiology	100	
Reflex		78
Transmission		67
Reception		56
EEG		22
CNS Physiology		11
Renal Physiology	89	
Urinalysis		78
Tubular Filtration		33
Hormonal Control		22
Muscle Physiology	100	
Skeletal		100
Smooth		55
EMG		22
Hematology	89	
Blood Type		89
Hb		89
Hematocrit		78
Clot Time		78
Histology		78
Sedimentation		33
Respiration	78	
Rate and Volume		78
Respiration Drive		55
Endocrinology	67	
Thyroid Hormone		56
Sex Steroids		56
Hormonal Bioassay		33
Epinephrine		22
Insulin		22
Nutrition	55	
Digestion		44
Absorption		22
Motility		11
Metabolism	55	
Calorimetry		55
Thermoregulation		22
Enzymes		22

Results and Discussion

Six of the ten major topics in human physiology were represented in all of the manuals examined and two other topics (nutrition and muscle physiology) were present in all but one manual (Table 1). Endocrinology and reproduction were the only human physiology topics covered by less than half of the manuals reviewed. The homogeneous nature of human physiology manuals transcended not only topic areas, but alos subtopics as well. Nine laboratory exercises (histology, hematocrit, blood type, blood pressure, reflexes, vision, audition, urinalysis, osmosis) were present in all human physiology manuals. Eight of these exercises have direct clinical applications and all nine are relatively inexpensive, easy to carry out, have a high success rate, and are illustrative of important physiological phenomena. Additionally, eight of these exercises utilize the human organism as the experimental subject which possesses obvious advantages, not the least of which is stimulating student interest in the functional properties of their own bodies. Other exercises found in a majority of human physiology manuals also involved relatively inexpensive instrumentation and clinical observations. These included blood clot time, heart sounds, taste reception, renal homeostasis, and spirometry. *In vitro* digestion and frog skeletal muscle contraction were the only two experiments which did not utilize human subjects but were present in most

of the manuals observed. These experiments are, however, inexpensive and illustrative of physiological principals which would be impossible to conduct on human subjects. Those experiments infrequently observed in the manuals involved expensive equipment acquisitions (electrophoretic apparatus, ergometer, physiological recorder, and calorimeter), more extensive animal care and handling (microcirculation, ciliary movement, cell respiration, sex steroids and pregnancy bioassay) or required complex and tedious methodologies, (observation of *in vitro* circulation, CO_2 measurement, karyotype production, accurate pH measurement and tedious bioassays). It is somewhat surprising that such experiments as hemoglobin determination, sedimentation rate, blood pH, olfaction, renal clearance, and respiratory sounds were not more widely distributed since they meet the major criteria of those found in all human physiology manuals (i.e., human subjects, little equipment, clinical application). Perhaps tradition and conformity are reflected in the manual authors' experiment selection within temporal constraints relative to limited student laboratory time. Human physiology manuals were largely restricted to human subjects, involved clinical data gathering (with clinical applications), were pedagogical in presentation, required a minimum of bioinstrumentation and were fairly standardized among all manuals. This must be considered a logical approach to laboratory instruction in human physiology since such courses generally involve underclassmen who are bound for clinical careers. Additionally, human physiology class sizes may be larger than general physiology courses necessitating financial frugality.

Laboratory experiments in general physiology manuals fit into nine major topic areas (Table 2). The topics cellular processes and reproduction were not present in general physiology manuals and the area metabolism was added. Experiments and subjects covered in general physiology manuals were much more heterogenous than those in human physiology manuals. This is exemplified by the fact that subtopics catagories were highly diversified and only three of the major topic areas were present in all general physiology manuals (circulation, neurophysiology and muscle physiology). The experiments common to all general physiology manuals (cardiac cycle, EKG, and skeletal muscle contraction) are all time tried experiments with a high level of success and clearly represent basic physiological phenomena. Probably for the same reasons as in human physiology manuals, the most commonly used experiments were the most inexpensive and utilized human subjects. However, there was a much greater tendency to utilize more sophisticated instrumentation (physiological recording systems with pleysmograph, EEG, EMG capability) than was the case in human physiology manuals. Additionally, there were more *in situ* investigations (direct blood pressure, blood volume, CNS physiology, tubular filtration, hormonal control, smooth muscle contraction, endocrinology, absorption, gastric motility, and calorimetry) which require animal maintenance and handling skills. In accordance with the theme of general physiology courses, these manuals also tended to offer a more varied array of experimental subjects (Table 3) although humans were again the most commonly utilized subject within five of the major topic areas.

Most human physiology experiments fit category 1 of independent student involvement while general physiology manuals exhibited a predominance of student involvement fitting categories 2 and 3 (Table 4). These results are consistent with the assertion that human physiology manuals tend to be clinically oriented where precision in technique is stressed and data are compared to previously determined ranges and averages. General physiology experiments involved much more data interpretation by the student, presumably to more completely develop an appreciation of the use of scientific method and design. Category 3 experiments

TABLE 3. *Percentage of General Physiology laboratory manuals using each experimental animal for each major topic area.*

Topic	Utilizing					
	Humans	Other Mammals	Reptiles	Amphibians	Fish	Invertebrates
Circulation	100	67	100	100	0	11
Endocrinology	11	56	0	0	0	0
Hematology	89	0	0	0	0	0
Metabolism	44	22	0	11	11	0
Muscle Physiology	22	44	0	100	0	0
Neurophysiology	78	22	0	78	0	0
Nutrition	0	33	0	0	0	11
Renal Physiology	55	22	0	22	22	11
Respiration	78	0	0	0	0	0

which involve the student directly in experimental design were totally lacking in human physiology manuals and surprisingly scarce in general physiology manuals (only two manuals included any category 3 experiments at all). Undoubtedly, category 3 experiments necessitate a reduction in the quantity of data collection and in the breadth of experimentation since student time is required in developing experimental design. The inclusion of at least one category 3 experiment would, however, present the student in physiology with first hand experience with the process of scientific method.

Appendix

Human Physiology

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3. EGAN, M.E. AND J. P. MCMILLAN. 1979. Human Function, A Laboratory Manual, W. B. Saunders Co. Philadelphia, PA 111 p.
4. FOX, S. I. 1980. A Laboratory Guide to Human Physiology Concepts and Clinical Applications. 2nd Ed. Wm. C. Brown Co., Dubuque, IA 290 p.
5. JACOB, S. W., C. A. FRANCONE AND W. J. LOSSOW. 1982. Laboratory Manual for Structure and Function in Man 5th Ed. W. B. Saunders Co. Philadelphia, PA 276 p.
6. KALBUS, B. H. AND K. G. NEAL. 1978. A Laboratory Manual for Human Physiology 2nd Ed. Burgess Pub. Co. Minneapolis, MN 399 p.
7. MCCLINTIC, J. R. 1976. Human Anatomy and Physiology. A Laboratory Manual. J. Wiley and Sons Inc. New York, NY 281 p.

TABLE 4. *Percentage of all laboratory exercises in each catagory of independent student involvement for human Physiology and General Physiology laboratory manuals (see text for description of catagories 1, 2, and 3).*

Manual Type	Category		
	1	2	3
Human Physiology	61.2	38.8	0.0
General Physiology	40.9	57.4	1.7

8. NICPON-MARIEB, E. 1981. Human Anatomy and Physiology Laboratory Manual. Benjamin/Cummings Pub. Co. Inc. Menlo Park, CA 216 p.
9. RUSSELL, G. K. 1978. Laboratory Investigations in Human Physiology. MacMillan Pub. Co. Inc. New York, NY 342 p.

General Physiology

1. BENSON, H. J., A. TALARO AND K. P. TALARO. 1982. Physiological Adaptations. Wm. C. Brown Co., Dubuque, IA 340 p.
2. COSTA, P. J. AND R. G. COTTY. 1980. Laboratory Textbook in Anatomy and Physiology, 4th Ed. Kendall/Hunt Pub. Co. Dubuque, IA 316 p.
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11. WITHERSPOON, J. D. 1970. The Functions of Life; A Laboratory Guide for Animal Physiology. Addison-Wesley Pub. Co. Reading, MA 210 p.

SOIL AND ATMOSPHERIC SCIENCES

Chairman: JOHN T. SNOW
Department of Geosciences
Purdue University, West Lafayette, Indiana 47907

Chairman-Elect: H. RAYMOND SINCLAIR
State Soil Scientist
410 Montrose Court, Indianapolis, Indiana 46234

ABSTRACTS

Soil Moisture Regimes in a Wooded Toposequence in Tippecanoe County. C. J. EVANS AND D. P. FRANZMEIER, Department of Agronomy, Purdue University, West Lafayette, Indiana 47907.——Characterization of soil moisture regimes provides one basis for a genetic interpretation of soil properties, so that soil morphological properties can then be used as indicators of the wetness characteristics of soil. In this way, gray (≤2 chroma) mottles in the soil have been used as indicators of soil saturation. However, the predictive value of gray colors is not quantitative. A soil zone which is periodically saturated may not always have gray mottles, and, conversely, the absence of gray color cannot always be interpreted as an absence of saturation.

Soil scientists have seen that, within a toposequence, the presence or absence of gray mottles will be related to slope position. It has not been certain, however, whether topography influences only the persistence of the water table itself, or whether it controls the other factors which must combine with saturation to cause gray mottles. The purpose of this study was to characterize the moisture regimes of soils in a toposequence. Three sites were selected in a woods where soils formed a loess over loamy glacial till—Miami soil in a backslope position, Fincastle soil in a swell position, and Treaty soil in a swale position. We measured water table levels, dissolved oxygen content of soil water, redox potential, and hydraulic conductivity.

Both slope position and gray colors predicted water table levels in a relative fashion along the toposequence; however, the Miami soil, which had no ≤2 chroma mottles, was occasionally saturated to within 55 cm of the soil surface. Length of saturation within 1.5 m of the surface also varied with slope—the Treaty soil was saturated 62.0% of the time, Fincastle was saturated 59.0% of the time, and Miami was saturated 32.0% of the time. Dissolved oxygen increased from swale to backslope. Low oxygen levels (< 5 ppm) occurred 43.0% of the time in the Treaty soil, 32.0% of the time in the Fincastle soil, and 12.0% of the time in the Miami soil. Expressed as a percentage of saturation time, oxygen levels less than 5 ppm occurred 69.0% of the time in Treaty, 54.0% of the time in Fincastle, and 38.0% of the time in Miami. Redox potential was monitored only at the Fincastle and Treaty sites, and Treaty had values less than +200 mV 54.0% of the time, while measurements in Fincastle were below +200 mV only 12.0% of the time. In contrast, Treaty had values greater than +600 mV 13.0% of the time, while 48.0% of the measurements at Fincastle were above +600mV. Hydraulic conductivity also trended along slope lines.

A Wind-Resource Prospecting Study for Indiana. STEPHEN A. JUSTHAM, TORU OTAWA, AND DAVID A. SCHOEN, Department of Geography, Ball State University,

Muncie, Indiana 47306.——The major objective of this study is to provide wind data requisite to planning for the placement of wind energy conversion systems (WECS) in the State of Indiana. Available wind data are usually recorded at locations such as weather stations and airports; therefore, the wind characteristics of intermediate areas are not well known due to the sparsity of data sources. This study provides a quantitative estimate of wind power potential for Indiana via a computer model. The model is based upon the theory of the conservation of mass and interpolates point-source wind data to produce statistically meaningful data for areas where wind recording stations do not exist. This approach will help WECS planners and users to pinpoint potential sites for the installation of meteorological instruments in order to obtain site-specific wind data.

Radar Signatures Associated with Damaging Straight Line Winds. RON W. PRZYBYLINSKI, National Weather Service, Indianapolis, Indiana 46204.—Approximately nine cases of severe weather occurring in the state of Indiana during the last three years (1980-81-82) have been examined and related to the bow echo configuration on radar. Similarities between the bow echo and damaging straight line winds have been noted. Case studies relate each of the occurrence of damaging winds in excess of 60 mph over scales ranging from only one convective cell to convective lines or clusters. Concentration on the recognition of the bow echo on radar during severe weather warning operations at the National Weather Service Forecast Office has resulted in small increases in warning lead time and decreases in the false alarm rate.

Objective Analysis of Mesoscale Disturbances Using Surface Meteorological Observations. DAVID R. SMITH AND JOHN T. SNOW, Department of Geosciences, Purdue University, West Lafayette, IN 47907.——Surface meteorological data can provide an important source of information concerning mesoscale disturbances due to the spatial (less than 100 km) and temporal (hourly) resolution of the data. For example, the systems responsible for severe convective activity can penetrate undetected through the synoptic upper air network. This makes the forecasting of severe weather very difficult. However, the frequency of the surface observations in a data dense region such as the Upper Midwest can provide important clues for areas of potential convective development some two to four hours in advance. Because of the large amount of data and the short time frame in which it must be processed and analyzed, a numerical objective analysis scheme is a necessity. The *P*urdue *R*egional *O*bjective *A*nalysis of the *M*esoscale (PROAM) is such a scheme. PROAM was especially designed to interpolate the randomly distributed surface observations onto a uniform grid in order to analyze variables (observed and computed) which help identify mesoscale disturbances.

Recent modifications to PROAM to improve its flexibility and performance to include a multiple iteration correcting technique, are discussed. Results involving both analytic distributions (to establish error limits) and actual observations (to demonstrate its utility in a nowcasting situation) are presented to demonstrate its effectiveness in identifying mesoscale disturbances.

Use of Sewage Sludge in Establishment of Northern Red Oak Seedlings on Adverse Sites. DOUGLAS M. STONE, North Central Forest Experiment Station, Bedford, Indiana 47421.——Northern red oak (*Quercus rubra* L.) was direct seeded on two acidic coal spoil areas in southern Ohio. Planting spots were prepared to reduce compaction and rock content; processed paper mill sewage sludge was used as a soil amendment. Treatments were: 1) scalping a 30 x 30 cm planting spot, 2)

boring a 30 cm diameter planting hole 30 cm deep, 3) planting hole + 10 l.(0.35 ft^3) of sewage sludge mixed with spoil material, and 4) planting hole + 20 l.(0.7 ft^3) of sludge. Treatments were applied to 14-tree plots replicated four times on each site. Four stratified acorns were sown at each planting spot and covered with 2-3 cm of spoil in early May 1982.

Spoil on one site was highly acid (pH 2.7); germination averaged 17.0%, growth and vigor of seedlings was low. The other site was less acid (pH 4.5); 25 to 95.0% of the planting spots had one or more living seedlings after the first growing season. Germination rate averaged 95.0% on scalped spots, 50.0% on spoil without sewage sludge; 66.0% on the spoil-sludge mixture and 25.0% with the high rate of sludge. Germination inhibition at the high sludge rate is attributed to nitrite toxicity. Revegetation of extremely acid spoil will require liming in addition to organic amendment. Incorporation of sewage sludge on moderately acid sites shows promise in establishment of red oak by direct seeding.

Potential of Amending Soil with Industrial Slags

P. N. Coody and D. W. Nelson
Department of Agronomy
Purdue University, West Lafayette, Indiana 47907

Introduction

Industries in the Great Lake States annually generate large quantities of waste materials that require disposal. However, many of the wastes can be recycled or used for productive purposes. For example, antibiotic production wastes, vegetable processing wastes, papermill lime sludges, and a variety of other industrial sludges have been added to soils to provide plant available nutrients and to increase organic matter levels (Nelson and Sommers, 1979); Simpson et al., 1979; Blondin et al., 1982; Smith and Peterson, 1982). Other wastes such as fly ash, cement kiln flue dusts, and blast furnace slags have been used as soil liming agents because they contain oxides, carbonates, or silicates of divalent alkaline earth metals (Tisdale and Nelson, 1956; Whittaker et al., 1959; Walker et al., 1979). A number of other industrial wastes (e.g., foundry slags, rockwool insulation wastes, and water softening waste slurries) may be potentially useful as liming materials (Goodwin et al., 1982). However, little is known about the effectiveness of most of industrial wastes in neutralizing soil acidity or about the phytotoxicity of the materials. The objective of this study was to determine the usefulness of two types of industrial wastes, foundry slags and rockwool manufacturing residuals, as soil liming agents through a combination of physical and chemical characterization and soil incubation studies. Since solubility (primarily particle size) and neutralizing potential (calcium carbonate equivalent) are important criteria in evaluating materials as liming agents, major emphasis was placed on physical and chemical characterization of wastes.

Materials and Methods

Samples of wastes were collected from manufacturing facilities in the Midwestern United States. The wastes were air-dried and the particle size distribution was determined by sieving. A subsample of each waste was ground to <0.5 mm and a portion of the ground sample was digested with $HF:HNO_3$ (Pratt, 1965). Calcium, Mg, Fe, Mn, Zn, and Cu concentrations in the digests were determined by atomic absorption and P and Si levels were determined by colorimetric methods (Sommers and Nelson, 1972; Kilmer, 1965). The calcium carbonate equivalent (CCE) was determined by equilibrating a sample of the ground waste with a known amount of 0.5 *N* HCl and backtitrating unreacted HCl with standard NaOH. The amount of HCl neutralized per unit weight of waste was expressed as a percentage of the HCl neutralized per unit weight of reagent grade $CaCO_3$.

Three moderately acidic surface horizons of Indiana soils were used in the incubation study. Maumee sandy loam (sandy, mixed, mesic Typic Haplaquoll) had the following characteristics: pH, 4.7; CEC, 18.1 meq/100 g; organic C, 1.64%; lime requirement, 10.4 Mtons $CaCO_3$/ha. Chalmers silty clay loam (fine silty, mixed, mesic Aeric Ochraqualf) had the following characateristics: pH, 5.9; CEC, 27 meq/100 g; organic C, 2.2%; lime requirement, 6.1 Mtons $CaCO_3$/ha. Russell silt loam (fine-silty, mixed mesic Typic Hapludalf) had the following characteristics (pH, 5.3; CEC, 21 meq/100 g; organic C, 1.6%; lime requirement, 7.5 Mtons $CaCO_3$/ha).

One hundred g samples of soil (<2 mm) were treated with variable amounts

of unground or ground (<0.86 mm wastes, reagent grade $CaCO_3$ (powder), or commercial agricultural limestone (28% was <0.23 mm) to provide from 1 to 4 times the lime requirement of the soil. The water content of the soil was maintained at -0.3 bar pressure 24% H_2O, w/w) and the samples were incubated at 24°C for up to 4 months. The pH of amended and unamended soils was periodically measured during the incubation by a glass electrode using a 1:1 soil:water ratio.

The phytotoxicity of wastes was evaluated by amending 1.5 kg samples of soil contained in plastic pots with rates of wastes or $CaCO_3$ similar to those used in soil incubation studies, adding adquate levels of N, P, and K to samples, and planting 4 corn or soybean seeds in each pot. Plants were grown in the greenhouse for 6 weeks at 26°C, the above ground portions of the plants were harvested, and dry matter yield per pot was determined.

Results and Discussion

Particle size analysis revealed that a small proportion (avg. = 6.5%) of foundry slag were >3.35 mm (>6 mesh) in diameter, whereas more than 30% of rockwool waste was >3.35 mm (Table 1). A high proportion (avg. = 61.1%) of slag particles had a diameter between 0.86 (20-mesh) and 3.35 mm, but only 23.4% of rockwool particles were in that size range. More than 46% of rockwool particles were < 0.86 mm, whereas an average of 32.4% of foundry slag particles were < 0.86 mm. From 25-35% of the particles in much of the agricultural limestone in Indiana are <0.23 mm (<60-mesh), whereas only 2.2% and 7% of slags and rockwool waste, respectively, were <0.23 mm. The low proportion of small particles (<0.23 mm) suggests that the wastes may have low solubility and, thus, be slow to react.

The CCE of rockwool waste and slags were 89 and 94.9%, respectively (Table 1). The Ca, Mg, and Si concentrations in the slags averaged 25.0, 0.5, and 19.3%, respectively, whereas the rockwool waste contained 13.7% Ca, 3.6% Mg, and 17.7% Si. The substantial amounts of Ca and Si in wastes suggest that the high CCE values originate largely from the presence of $CaSiO_3$ and CaO (the pure form of these compounds have CCE values of 86 and 179%, respectively). The rockwool waste may also contain MgO and/or $MgSiO_3$ (both good liming agents). The high CCE value of the wastes suggest that they may be useful as soil liming agents. The slags and rockwool waste contained 2.1 and 5.1% Fe, respectively, and trace amounts of Mn, Zn, Cu, and P (data not presented).

The addition of high rates (>11.4 Mton/ha) of the four slags resulted in modest increases in soil pH (Table 2). After 2 months of incubation the increases of pH obtained from addition of unground slag were 20% of those observed with similar

TABLE 1. *Physical and chemical characteristics of industrial wastes used in the study (all data are expressed on dry weight basis).*

Component	Slag 1	Slag 2	Slag 3	Slag 4	Rockwool waste
Particle size dist.					
>3.35 mm, %	7.6	2.1	10.9	5.3	30.3
0.86-3.35 mm, %	54.5	57.5	71.3	61.1	23.4
<0.86 mm, %	37.9	40.4	17.8	33.6	46.3
<0.23 mm, %	3.7	2.1	0.8	2.1	7.0
$CaCO_3$ equiv., %	91.5	109.5	89.5	89.0	89.0
Ca, %	25.0	25.5	22.5	26.8	13.7
Mg, %	0.6	0.4	0.4	0.5	3.6
Si, %	16.4	17.7	22.8	20.2	17.7

TABLE 2. *Increase in pH of two soils as affected by addition of variable rates of calcium carbonate, limestone, or slags.*

Material added		1 month incubation	2 month incubation	
Type	Amount	Maumee sl.+	Chalmers sicl.+	Maumee sl.+
	Mtons/ha	increase in pH		
$CaCO_3$	5.7	1.01	1.48	1.39
$CaCO_3$	11.4	1.75	1.67	2.02
$CaCO_3$	17.0	1.94	1.76	2.51
$CaCO_3$	22.7	2.09	1.78	2.69
Limestone	11.4	0.56	0.98	0.82
Limestone	22.7	0.95	1.09	1.11
Slag 1 (Orig.)	22.7	0.14	0.28	0.20
Slag 1 (<0.86 mm)	11.4	0.47	0.48	0.47
Slag 1 (<0.86 mm)	22.7	0.59	0.68	0.70
Slag 2 (Orig.)	22.7	0.19	0.19	0.21
Slag 2 (<0.86 mm)	11.4	0.20	0.25	0.38
Slag 2 (<0.86 mm)	22.7	0.28	0.36	0.50
Slag 3 (Orig.)	22.7	0.11	0.16	0.26
Slag 3 (<0.86 mm)	11.4	0.15	0.15	0.31
Slag 3 (<0.86 mm)	22.7	0.19	0.16	0.25
Slag 4 (Orig.)	22.7	0.13	0.23	0.22
Slag 4 (<0.86 mm)	11.4	0.21	0.19	0.30
Slag 4 (<0.86 mm)	22.7	0.31	0.27	0.38

+Lime requirements for Maumee sandy loam and Chalmers silty clay loam were 10.4 and 6.1 metric tons of $CaCO_3$ per hectare, respectively.

rates of limestone and only 8% of those attained from application of $CaCO_3$. Grinding slag to <0.86 mm increased the solubility such that addition of 22.7 Mtons/ha raised the pH of Maumee soil to 41% and 17% of that attained from limestone and $CaCO_3$, respectively. Similar results were attained with slag addition to Chalmers soil. The increase in soil pH observed during the period between 1 and 2 months of incubation suggests that the pH may have increased further if the incubation had continued. A number of studies with agricultural limestone have suggested that about 50% of the long term pH increase occurs in the first 2 months of incubation. The pH increase obtained with limestone was 50 and 41% of that with $CaCO_3$ (11.4 Mtons/ha rate) added to Chalmers and Maumee soil, respectively, indicating the effect of particle size on rate at which acidity is neutralized.

Calcium carbonate completely reacted in Russell silt loam soil during 8 weeks of incubation (Table 3) After 16 weeks of incubation unground rockwool waste (all rates) increased soil pH to 35% of that obtained with a 7.5 Mtons/ha addition of $CaCO_3$. The pH levels of soil treated with rockwool waste increased with increasing time of incubation suggesting that given enough time the waste may be a satisfactory liming agent. The rate of reaction of ground rockwool waste was faster than that of unground waste. After 16 weeks of incubation ground rockwool waste added to soil at rates of 8.4, 16.9, and 25.3 Mtons/ha increased pH to 38, 48, and 59% of that obtained from adding 7.5 Mtons of $CaCO_3$/ha.

Greenhouse experiments established that similar dry matter yields of corn and soybeans were obtained in soils treated with high rates (>20 Mtons/ha) of wastes plus NPK fertilizer and soils receiving only NPK (data not shown). However,

TABLE 3. *Increase in pH of Russell silt loam soil as affected by addition of varying rates of calcium carbonate or rockwool waste (soil had a lime requirement of 7.5 metric tons/ha).*

Material added		Incubation time, weeks			
Type	Amount	2	4	8	16
	Mtons/ha	increase in pH			
$CaCO_3$	7.5	0.79	1.94	2.01	2.01
	15.0	0.98	1.93	2.10	2.17
	22.5	1.13	1.98	2.31	2.32
Rockwood waste (Orig.)	8.4	0.05	0.10	0.37	0.70
	16.9	0.16	0.27	0.42	0.72
	25.3	0.23	0.42	0.43	0.72
Rockwool waste (ground)+	8.4	0.05	0.32	0.49	0.77
	16.9	0.24	0.50	0.64	0.97
	25.3	0.52	0.62	0.97	1.19

+Rockwool waste was ground such that 89% was <0.5 mm.

the NPK treatment gave higher yields than the unfertilized control. This finding indicates that slags and rockwool waste were not phytotoxic to plants and that it is likely these wastes can be safely applied to cropland.

The results of this study indicate that foundry slags and rockwool waste may be useful as soil liming agents if they are applied at high rates and long periods of time are allowed for neutralization of soil acidity. Grinding the wastes increases solubility and speeds up the rate at which soil acidity is neutralized.

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Conservation Tillage Studies on a Clermont Silt Loam Soil

E. J. KLADIVKO, D. R. GRIFFITH, and J. V. MANNERING
Department of Agronomy, Purdue University, West Lafayette, Indiana 47907

Introduction

Conservation tillage systems have become increasingly popular as a means for a farmer to save time and fuel in the preparation of land for planting, as well as for reduction of soil erosion. It is important that an appropriate tillage system be chosen for a given soil type, since tillage techniques vary in their adaptability to different soils. Studies at Purdue have resulted in some guidelines for choosing a tillage system to fit a particular soil (Galloway et al., 1981), but more work is needed on soil properties and crop growth on "problem" soils, as they are affected by tillage.

The Clermont soil occurs extensively in southeastern Indiana and southwestern Ohio. Its morphology and hydrology have been described by King and Franzmeier (1981). The Clermont silt loam is a Typic Ochraqualf with a 0-2.0% slope, about 73.0% silt, 10.0% clay, and less than 1.0% organic carbon in the plow layer, and a fragi-like pan at a depth of about 3 feet. The soil has several problems which make it difficult to manage. 1) The soil is highly erosive in spite of the flat to gently sloping topography, due to the high silt and low organic matter contents. Eroded rills and channels could be observed even on gentle slopes at the farm. 2) Trafficability is usually poor in the spring, due to wetness problems, which results in delayed planting and reduced yield potential. The wetness is due to a seasonally high water table and to the slow hydraulic conductivity of the plow pan. 3) Rooting depth can be restricted by the excessive spring wetness. This can result in aeration problems in the spring and drought problems in mid-summer.

It was hypothesized that with appropriate tillage techniques, the management problems on the Clermont soil could be reduced. The objective of this experiment is to determine the adaptability of varous tillage systems to the Clermont soil, with respect to (1) early crop growth, crop yield, and rooting patterns, and (2) soil physical properties, including soil stability against erosion, soil temperature and water content, trafficability, and hydraulic conductivity. The results of measurements taken during the first three years of this long-term study are discussed here.

Experimental Treatments and Methods

Four tillage treatments and two crop rotation treatments were replicated four times in a randomized block design. Tillage treatments were spring moldboard plow and disk twice, fall chisel and disk twice in spring, spring disk twice, and no-till. Plow, chisel, and disk plots were cultivated once. Crop rotations were continuous corn and corn-soybeans. Seeding rate was 24,000 seeds/acre for corn and 124,000 seeds/acre for soybeans (calculated from actual measure of 48 lbs/acre, with data from Wilcox, et al., 1981).

Stands were not significantly affected by tillage except for no-till beans in 1980 which had a severe infection of phytophthora root rot. Corn stands averaged 21,900 and 20,400 plants per acre for 1980 and 1981 respectively, while soybean stands averaged 8.1 and 6.6 plants per foot in the same years. Planting dates for corn were June 9, 1980, June 17-18, 1981, and April 28-29, 1982, and for soybeans were June 10, 1980, June 18-19, 1981, and May 20, 1982. Weed control was adequate on all plots.

Percent surface residue cover was estimated by a photographic grid technique within a day after planting. A rectangular frame (76 cm x 51 cm) is placed on the ground and centered over a row, such that the frame extends 38 cm on either side of the row to the row middles (76 cm row spacing used in planting). A color slide is then taken from a nearly vertical angle over the soil and frame. The slides are projected onto a gridded screen with the same relative dimensions as the frame. The number of intersections of residue pieces with the 50 grid marks on the screen is counted and multiplied by 2 to get percent surface cover. Three adjacent rows in the center of each plot were photographed and counted to obtain an average value for each plot.

Size distribution of water-stable aggregates was determined by the Yoder sieving procedure, as described by Kemper and Chepil (1965). In this procedure, aggregates between 2mm and 8mm in diameter are oscillated on a nest of 4 sieves under water for 10 minutes, and the weight of aggregates remaining after sieving is determined. Soils which are more water-stable will have more large aggregates remaining after sieving. An average diameter is calculated based on the weight of soil in each size class rather than on the number of aggregates in each class. The calculation of this mean weight diameter (MWD) is used to reflect the thought that a few large aggregates are more important than many small aggregates in determining the resistance of surface soil to raindrop detachment and erosion.

The data were statistically analyzed using analysis of variance tests. Duncan's multiple range test (Steel and Torrie, 1960) was then performed (95.0% confidence level) on those data where tillage was a significant treatment effect.

Results and Discussion

Residue cover and size distribution of water stable aggregates

Percent surface residue cover remaining after planting (Table 1) showed the typical trend of more cover with less intensive forms of tillage. Plowing buried esentially all crop residues, no-till systems left the surface nearly totally covered, and chisel and disk systems were intermediate. Cover was less following soybeans across all tillage methods, due to the lower production of residue by soybeans than by corn. Two-way analysis of variance showed that the effects of tillage, previous crop, and the interaction of tillage and previous crop were all highly significant (99.0% confidence level).

The concentration of residues near the soil surface with reduced tillage protects the soil surface from raindrop inpact and aggregate disruption, and also supplies organic matter for microbial production of stable aggregates. Mean weight diameter of water stable aggregates, measured immediately prior to planting, showed a significant trend of larger stable aggregates with reduction in tillage

TABLE 1. *Percent surface residue cover measured shortly after planting, 1982.*

	Previous Crop	
Tillage	Corn	Soybeans
Plow	0a†	0a
Chisel	12b	2ab
Disk	17c	3b
No-till	96d	87c

†Values in the same column followed by the same letter were not statistically different at the 5.0% level.

TABLE 2. *Size distribution of water-stable aggregates (0-7.5 cm depth) prior to planting, April 28, 1982.*

Tillage	Mean Weight Diameter (mm) Rotation		
	CC[1]	BC[2]	CB[3]
Plow	0.78 a[4]	0.68 a	0.74 a
Chisel	0.97 a	0.60 a	0.91 ab
Disk	0.95 a	1.34 b	1.15 b
No-till	0.73 a	1.18 b	1.10 ab

[1]continuous corn
[2]corn in 1982, following 1981 soybeans
[3]soybeans in 1982, following 1981 corn
[4]values in the same column followed by the same letter were not statistically different at the 5.0% level.

intensity, especially in the soybeans-corn and corn-soybeans rotation plots (Table 2). Although there was a significant increase in residue cover (Table 1) and stable aggregate size (Table 2) with reduction in tillage intensity, there was no significant correlation between cover and aggregate size at the April sampling date. The smaller aggregate size in the no-till plots compared with the other tillage treatments in continuous corn is thought to be due to wetter soil conditions which could have caused aggregate breakdown during sample preparation.

Stable aggregate sizes measured at mid-summer (Table 3) were greater than those measured in April for all treatments, showing the typical increase in aggregate size and stability during the first half of the growing season, due to microbial decomposition of crop residues. The trend of increased aggregate size with reduced tillage intensity was even more evident in July than in April. No-till plots generally had the largest stable aggregates due to the high concentration of residues near the surface. Even the chisel systems, which incorporate residues vertically into the soil to depths of about 4-5 inches, left enough residues near the surface to improve stability. Surface residue cover prior to planting and July stable aggregate size were significantly correlated (99.0% level), with a correlation coefficient of 0.66.

At the July sampling date there was also a significant effect of crop rotation on aggregate size, with the soybean plots having smaller aggregates than the two rotations that were growing corn. This more erosive soil condition under soybeans has often been observed by farmers and researchers but is not yet understood.

TABLE 3. *Size distribution of water-stable aggregates (0-7.5 cm depth), July 26, 1982.*

Tillage	Mean Weight Diameter (mm) Rotation		
	CC[1]	BC[2]	CB[3]
Plow	1.49 a[4]	0.91 a	0.87 a
Chisel	1.76 ab	1.82 b	1.39 b
Disk	2.13 bc	1.98 b	1.87 c
No-till	2.33 c	2.69 c	2.33 c

[1]continuous corn
[2]corn in 1982, following 1981 soybeans
[3]soybeans in 1982, following 1981 corn
[4]values in the same column followed by the same letter were not statistically different at the 5.0% level.

TABLE 4. *Plant height (in) 8 weeks after planting, 1981.*

	Corn Height		Soybean Height
	Prior crop		
	corn	soybeans	corn
Plow	101 b†	101 a	30 a
Chisel	103 b	99 a	32 b
Disk	101 b	100 a	32 b
No-Till	93 a	95 a	30 a

†Values in the same column followed by the same letter were not statistically different at the 5.0% level.

Crop growth and yields

Plant height 8 weeks after planting was measured as an index of early growth rates (Table 4). Corn heights were lower in no-till plots than in plow, chisel, and disk plots, probably due to the cooler soil temperature caused by heavy residue cover (temperature data not shown). The reduced growth rate was more significant for continuous corn than for corn following soybeans due to the difference in residue cover following these two crops. Soybean heights were significantly affected by tillage, but not in any consistent trend with tillage intensity.

Two-way analysis of variance (95.0% level) showed corn yields to be significantly affected by tillage in 1981, by previous crop in 1982, and by the interaction of tillage and previous crop in 1980, 1981, and 1982 (Table 5). The effect of tillage on yield was primarily due to lower yields with no-till systems compared to other tillage systems under continuous corn culture. When corn followed soybeans, there was no significant effect of tillage on yield. The significant interaction term indicates that adaptability of different tillage systems becomes more important under continuous rowcrop culture, or conversely, that the choice of tillage system is less critical under rotation systems. In 1982, corn yields following soybeans were significantly greater than continuous corn yields, indicating some type of positive rotation effect.

Soybean yields (Table 6) were not significantly affected by tillage in any of the three years of the study, except for no-till plots in 1980. No-till yields during 1980 were very low due to a large amount of Phytophthera root rot, which was minimized in subsequent years by planting a resistant variety.

Summary and Conclusions

1. No-tillage and shallow tillage over a 3-year period have increased the water-stable aggregate size of the surface of the Clermont silt loam.

TABLE 5. *Corn yields (bu/acre), 1980-1982.*

	Previous Crop					
	Corn			Soybeans		
Tillage	1980	1981	1982	1980	1981	1982
Plow	114	123 b†	187	116	122 a	197
Chisel	122	131 b	194	112	119 a	187
Disk	117	125 b	181	120	120 a	196
No-Till	105	105 a	160	120	116 a	197

†Values in the same column followed by the same letter were not statistically different at the 5.0% level.

Table 6. *Soybean yields (bu/acre), 1980-1982.*

	Previous Crop		
		Corn	
Tillage	1980	1981	1982
Plow	39	43	52
Chisel	40	41	51
Disk	40	39	52
No-till	19*	42	49

*Phytophthera root rot reduced yield.

2. Percent surface residue cover after planting was greatest for no-till, intermediate for disk and chisel systems, and lowest for plow systems. Cover was greater following corn than following soybeans.
3. Early corn growth rates were slower in no-till systems than plow, chisel, and disk systems.
4. Corn yields were depressed in no-till treatments following corn, but were similar to conventional systems when corn followed soybeans. Soybean yields were unaffected by tillage treatment when Phytophthora root rot was not a problem.

Results from the first three years of this study indicate that no-till may be a viable management system for the Clermont soil, when used in a corn-soybean rotation. Yields of both corn and soybeans are comparable with other tillage systems, when weeds are controlled and when Phytophthora-resistant soybean varieties are used. Surface soil stability has also been increased by leaving crop residues near the soil surface, and this may ultimately help reduce erosion and improve the overall structure of the Clermont soil.

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Some Aspects of the Persistence Between Fall and Winter Mean Temperatures in Indiana

HUO-JIN HUANG and ROBERT F. DALE
Departments of Geosciences and Agronomy
Purdue University, West Lafayette, Indiana 47907

Introduction

Accurate predictions of winter mean temperature would be helpful in allocating fuel supplies to meet the anticipated energy demands for heating. While there has been little accuracy in deterministic long-range weather forecasts, persistence of temperature anomalies from one season to the next may provide some help in planning.

The day-to-day continuity of the synoptic weather provides the concept of persistence, which is important in synoptic weather prediction (MacDonald and Shapiro, 1964). Seasonal mean temperatures may or may not demonstrate this kind of persistence, however, because of the longer period involved. van Loon and Jenne (1975) used the persistence between adjacent seasons to estimate seasonal mean temperatures. They found that the highest correlation between two seasons occurred if the first were extremely cold or extremely warm.

The objective of this study is to examine, with correlation-regression methods and with a contingency table approach, the degree of persistence of temperature anomalies from fall to winter, using the climatological division mean temperature for fall and the following winter in Indiana.

Data and Procedures

The fall and the winter mean temperature for nine climatological divisions in Indiana were obtained from the monthly Climatological Data, Indiana (USDC, 1931-1980), from September to February, 1931-1980. The average mean temperatures for the three months, September, October, and November, represent the fall mean temperatures, and the averages of the three months, December, January, and February, the winter mean temperatures.

The divisional mean temperatures were also used to obtain the means for three larger regions and the State. The northern region (NORTH) included the Northwest (NW), North Central (NC) and Northeast (NE) divisions; the central region (CENTRAL) included the West Central (WC), the Central (CC) and East Central (EC) divisions; and the southern region (SOUTH) included the Southwest (SW), South Central (SC) and Southeast (SE) divisions. The state average temperature was computed as the average of the mean temperatures of the nine divisions (equal-weighting).

The correlation coefficient (r) was calculated as $r = \frac{s_{xy}}{s_x s_y}$, where s_{xy} is the covariance of the fall and the winter mean temperatures, and s_x and s_y are the standard deviations of the fall and the winter mean temperatures, respectively. An analysis of variance and an F-test (Panofsky and Brier, 1965, p. 93) was conducted to test the null hypothesis that $r = 0$ (no persistence).

To detemine if there were any temperature trend, the fall and the winter mean temperatures were each regressed on years. To determine the correlation between seasons, the winter mean temperatures were regressed on fall temperatures for each division and region and the State. The simple linear regression model, $Y = b_0 + b_1X$, was used, where Y is the dependent variable and

X is the independent variable, year in the trend analysis and fall mean temperature in the persistence study. Finally, contingency tables of winter on fall mean temperature anomalies were prepared, using three categories determined by plus and minus one-half standard deviation(s) about the respective means.

Results and Discussion

The temperature means and the standard deviations for the fall and the winter seasons for each of the nine divisions, three regions, and the state of Indiana for the period 1931-1980 are shown in Table 1. The covariances between fall and following winter seasons are also shown. The divisional mean temperatures for the fall range from 52.8°F in the NE to 57.4 in the SW, and for the winter from 26.9°F to 33.6.

The fitted regression coefficients for the time trend in fall and winter mean temperatures are shown in Table 2. The trend for the Central division is shown in Figure 1. All divisions showed a cooling trend but that for the Central division was the greatest. Undoubtedly, some of this cooling in Figure 1 can be attributed to changes in time of observation and network configuration, as shown by Nelson *et al.* (1979). This "non-climatic cooling", however, should not affect the persistence of adjacent fall and winter mean temperatures. For all regressions in Table 2 and Figure 1, winter showed a greater cooling trend than fall. The cooling trend for the State is shown as -0.03°F per year in the fall and -0.11°F per year in the winter. The trend regressions were signficant at the $\alpha = 0.10$ level in all divisions except for the NW in the fall. Since the time trends are significant, this may affect the evaluation of the true relationship between fall and winter mean temperatures, because the relation between the two time (or climate) trends may conceal the relation between the two time series. Therefore, the time trend was removed from the published means by subtracting the trend values from the mean temperature, or using departures from a "moving normal". Hereafter, the original mean temperatures will be referred to as "with-trend", and the departures from trend as "without-trend" cases.

TABLE 1. *Fall and the winter mean temperatures (°F), standard deviations, and covariances between fall and winter mean temperatures for 9 divisions, 3 regions and State of Indiana.*

	FALL (1931-1980)		WINTER (1931-1980)		
Division	Mean	Standard deviation	Mean	Standard deviation	Covariance
NW	53.47	2.07	26.87	3.56	1.52
NC	53.00	2.07	27.14	3.59	1.64
NE	52.81	2.08	26.88	3.57	1.97
WC	54.80	2.12	29.44	3.90	2.55
CC	54.36	2.01	29.56	3.92	2.52
EC	53.38	1.99	28.77	3.96	2.19
SW	57.39	2.03	33.64	3.62	1.71
SC	56.42	2.06	33.30	3.66	2.14
SE	56.70	1.96	33.58	3.56	1.74
Region					
NORTH	53.27	2.03	26.96	3.59	1.18
CENTRAL	54.43	1.99	29.33	3.88	1.78
SOUTH	57.09	1.96	33.56	3.68	1.24
State					
INDIANA	54.91	1.97	29.95	3.64	1.37

TABLE 2. *Regression coefficients for trend model, $Y = b_0 + b_1X$, where Y is either fall or winter mean temperature and X is year, r correlation coefficient, and F for testing significance of regression trend for 9 divisions, 3 regions and the State of Indiana. $F_{1,48;0.90} = 2.82$.*

Division	FALL(Y) on YEAR(X)				WINTER(Y) on YEAR(X)			
	b_0	b_1	r	F	b_0	b_1	r	F
NW	113.0	-0.031	-0.214	2.304	223.5	-0.101	-0.411	9.756
NC	122.5	-0.036	-0.250	3.200	237.0	-0.107	-0.437	11.330
NE	123.2	-0.036	-0.252	3.255	237.1	-0.108	-0.439	11.459
WC	153.2	-0.050	-0.347	6.571	304.6	-0.141	-0.527	18.457
CC	174.1	-0.061	-0.445	11.852	305.0	-0.141	-0.523	18.073
EC	125.0	-0.037	-0.270	3.774	296.0	0-137	-0.504	16.345
SW	133.6	-0.039	-0.281	4.115	234.5	-0.103	-0.415	9.987
SC	156.1	-0.051	-0.360	7.147	262.1	-0.117	-0.467	13.388
SE	129.0	-0.037	-0.275	3.927	228.1	-0.100	-0.408	9.586
Region								
NORTH	97.1	-0.022	-0.161	1.277	233.6	-0.106	-0.429	10.826
CENTRAL	113.7	-0.030	-0.222	2.488	291.5	-0.134	-0.504	16.345
SOUTH	104.0	-0.024	-0.179	1.589	233.7	-0.102	-0.419	10.221
State								
INDIANA	105.6	-0.026	-0.192	1.837	253.0	-0.114	-0.456	12.601

The correlations between fall and winter mean temperatures, both with- and without-trend, for the nine divisions, three regions and the State of Indiana are shown in Table 3. The r values are comparable to those of van Loon and Jenne (1975). The F values included in Table 3 for the "with-trend" case show that correlation of the fall and the winter mean temperatures are significant at the $\alpha = 0.10$ level for the Northeast, West Central, Central, East Central, South Central and Southeast divisions. Although these r values are not very high, they are all positive, suggesting some degree of persistence. In fact, 4% to 10% (r^2)

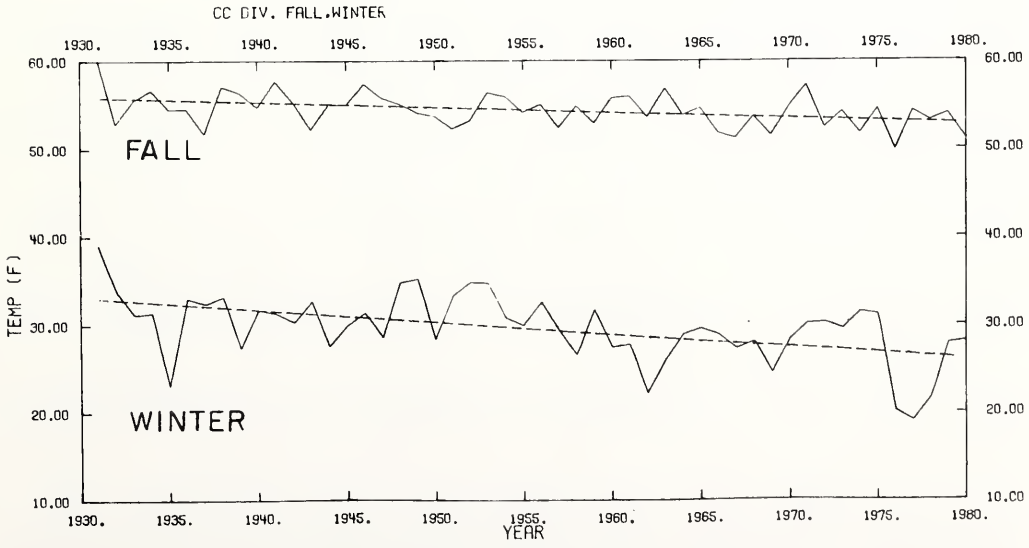

FIGURE 1. *Mean temperatures for fall and winter on years and linear trends (dashed lines) for the Central (CC) climatological division, Indiana, 1931-1980.*

TABLE 3. *Regression coefficients for model, $Y = b_0 + b_1X$ where Y is winter mean temperature and X is preceding fall mean temperature, correlation coefficient, and F value, with-and without-trend, for 9 divisions, 3 regions and the State of Indiana. $F_{1,48;0.90} = 2.82$.*

Division	WITH-TREND WINTER(Y) on FALL(X)				WITHOUT-TREND WINTER(Y) on FALL(X)			
	b_0	b_1	r	F	b_0	b_1	r	F
NW	7.990	0.353	0.206	2.127	0.018	0.212	0.132	0.851
NC	6.917	0.381	0.220	2.441	0.002	0.203	0.127	0.787
NE	2.842	0.455	0.265	3.625	-0.004	0.285	0.179	1.594
WC	-1.757	0.569	0.309	5.067	-0.001	0.260	0.157	1.197
CC	-4.429	0.625	0.320	5.476	0.000	0.213	0.115	0.639
EC	-0.617	0.551	0.279	4.052	0.000	0.296	0.165	1.348
SW	9.434	0.422	0.234	2.781	0.004	0.229	0.135	0.895
SC	5.089	0.500	0.284	4.211	0.000	0.230	0.138	0.930
SE	7.685	0.457	0.249	3.173	0.030	0.273	0.158	1.234
Region								
NORTH	11.641	0.288	0.162	1.294	0.002	0.167	0.103	0.516
CENTRAL	4.952	0.448	0.230	2.681	-0.001	0.239	0.138	0.928
SOUTH	15.048	0.324	0.178	1.571	-0.006	0.194	0.116	0.652
State								
INDIANA	10.561	0.353	0.191	1.817	0.000	0.202	0.121	0.708

of the variance in the winter mean temperatures is associated with the previous fall mean temperatures. However, when the trend effect is removed, none of the regressions are significant, and only 1% to 3% of the winter mean temperature variance is associated with fall temperatures. Only the fall-winter relationships of the Central division are presented in Figure 2, because of the similarity of

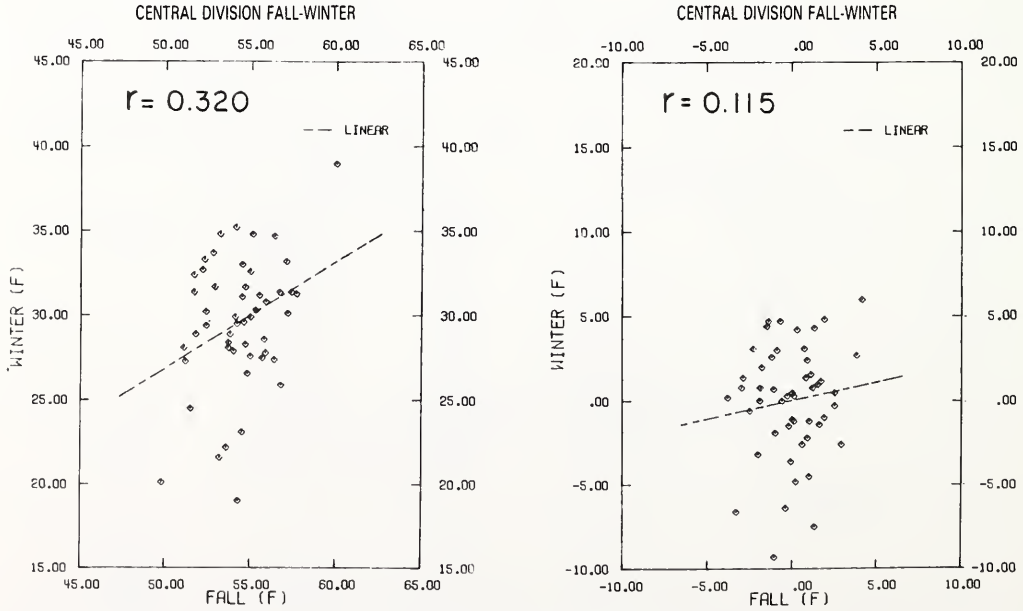

FIGURE 2. *Scatter diagram of winter on fall mean temperatures for the Central (CC) division for 1931-1980, and linear regression (dashed line), with-trend (left), and without-trend (right).*

the other data sets. The scatter diagrams show the reduction in the b_1 and r values from the with-trend to without-trend cases.

The contingency tables for the Central division fall and winter temperature quantiles for both with-trend and without-trend cases are shown in Table 4. For example, for the with-trend case, when the fall mean temperatures averaged below normal (less than $\bar{x}$ - 0.5s). two of the following winters also averaged below normal, six near normal and five above normal. The x^2 value was computed as $x^2 = \frac{(2-2.1)^2}{2.1} + \frac{(6-7.1)^2}{7.1} + \ldots \frac{(3-3.9)^2}{3.9} = 2.80$ which is tested against $x^2_{.10}$ with 4 degrees of freedom. The computed Chi-square values for each division, region and State are shown. It is concluded that the null hypothesis (that fall and winter mean temperatures are independent) can not be rejected, especially for the without-trend cases. This is consistent with findings using the correlation-regression method.

TABLE 4. *Contingency tables for winter on fall mean temperature anomalies for Central division and Chi-square values from similar contingency tables for nine devisions, 3 regions and the State of Indiana, with and without-trend. Values in the parentheses are expected frequencies.*
$\times^2_{4;0.90} = 7.78$. *B: below mean* ($\bar{x}$) – ½ *standard deviation* (s_x);
N: between $\bar{x}$ – ½ s_x *and* $\bar{x}$ + ½ s_x; *A: above* $\bar{x}$ + ½ s_x; *and*
T: total number of occurrences.

		WITH-TREND WINTER					WITHOUT-TREND WINTER			
		B	N	A	T		B	N	A	T
FALL	B	2 (2.1)	6 (7.3)	5 (3.6)	13	B	4 (4.1)	7 (3.6)	6 (4.3)	17
	N	5 (3.7)	12 (12.3)	6 (6.4)	23	N	4 (8.2)	8 (7.2)	3 (8.6)	15
	A	1 (2.2)	10 (7.8)	3 (3.9)	14	A	4 (4.8)	9 (4.2)	5 (5.0)	18
	T	8	28	14	50	T	12	24	14	50

Division	Chi-square value	Division	Chi-square value
NW	1.95	NW	0.71
NC	0.35	NC	0.20
NE	3.26	NE	0.96
WC	1.54	WC	0.08
CC	2.80	CC	0.19
EC	3.39	EC	0.36
SW	3.57	SW	0.13
SC	2.83	SC	0.22
SE	5.53	SE	0.55
Region			
NORTH	3.92	NORTH	0.64
CENTRAL	0.70	CENTRAL	0.06
SOUTH	2.50	SOUTH	0.21
State			
INDIANA	2.75	INDIANA	0.18

Summary

The persistence between fall and winter mean temperatures is not significantly high in Indiana. The trend analysis shows climatic cooling, with winter cooling more than fall. The "without-trend" analysis is believed to be a more represen-

tative evaluation of the persistence effect, since the existence of trend in both data sets conceals the true persistence relationship.

The fact that all correlations between fall and winter are positive indicates some persistence between fall and the winter mean temperatures in Indiana, but the correlation is too low to rely on persistence of temperature anomalies from fall to predict winter mean temperature. With the cooling trends in both time series, correlation of the published data, instead of the departures from trend, probably adds to the measure of persistence. It is suggested that for the short climatological records ($\leq$50 years) departures from trend should be used to obtain a more realistic evaluation of the temperature persistence between adjacent seasons.

Acknowledgment

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A Methodology for Considering the Effect of Weather on the Response of Corn Yields to Added Fertilizer: A Case Study in Indiana with Nitrogen

MICHAEL A. MACH and ROBERT F. DALE
Purdue University, West Lafayette, Indiana 47907

Introduction

Many studies have shown that that crop yield response to applied fertilizer is greatly influenced by weather and climate (Dale and Shaw, 1964; Voss, Hanway, Fuller, 1970; Bondavalli, Colyer, and Kroth, 1970; Isfan, 1979; and Shaw, Ross, and Meyers, 1980). Yet, in most agricultural experiments, weather is rarely considered quantitatively in the analysis and interpretation of results. If the experiment has been conducted over several years and/or locations, the cumulative effect of weather on yield has been pooled with the effects from other unexplained sources of variation and usually attributed in analyses of variance to Years and/or Locations, and their interactions with Treatments. Rising costs of crop production, especially in nitrogen fertilizers (Hargett and Berry, 1980), and decreasing returns to the farmer dictate the need for quantifying the interactions between various management and weather variables affecting crop production to allow for more intelligent management decisions.

The three most important weather variables affecting crop growth and final yields are light, temperature, and plant moisture (Cowan and Milthorpe, 1968). Coelho and Dale (1980) considered all three weather variables on each day, as they affect corn (Zea mays L.) growth, in a single environmental product, called the Energy-Crop Growth (ECG) variable.

$$ECG = (SR/L)(FLAI)(ET/PET)(FT) \quad (1)$$

SR is the daily solar radiation or short wave energy received on a horizontal surface, L is the latent heat of vaporization of water, LAI is the corn leaf area index, and FLAI is 1-exp(-0.79 LAI), used to convert the SR to that intercepted by the corn canopy. ET/PET is the ratio of actual to potential evapotranspiration, and FT is a temperature function reflecting the growth rate of corn relative to the maximum obtained from 28 to 32°C. Except for SR and L, all factors in (1) range from 0 to 1, which puts the units of ECG into cm of water evaporated if all of the intercepted short wave energy were converted to latent heat.

Coelho and Dale (1980) also found that FT alone, computed from the daily maximum and minimum temperatures and summed from date of planting (ΣFT), provided a slighly better estimate of the experimental corn ontogeny than that obtained using modified (10-30°C cutoff) growing degree days. For example, at ΣFT = 3 about 50% of the corn plants emerged, at ΣFT = 13 the growing point of the corn plants were rising above the soil surface, and at ΣFT = 37, about 50% of the corn plants were silked. By summing the daily ECG (1) over several different vegetative growth and grain filling periods, Mach (1982) found that the sum of the daily ECG from the date ΣFT = 13 (39 to 45 days before silking) to 50 days past silking provided the energy-crop growth index (ΣECG) most highly correlated with final grain yields. The basis for the ECG is that with ample solar radiation upon a corn canopy which completely covers the soil surface (FLAI = 1), with no plant moisture stress (ET/PET = 1), and with temperature at the

optimum for plant growth (FT = 1), the corn will make excellent growth. The greater the ΣECG, the greater will be the corn growth and final yield.

The objective of this paper is to demonstrate the use of ΣECG to quantify the effect of weather on the response of corn to added nitrogen fertilizer. To do this, we have used data from a long-term nitrogen (N) fertilizer field trial with corn evaluating the yield responses for selected corn hybrids and application methods.

Materials and Methods

A long term N fertility management trial was conducted by Dr. Stanley Barber and Mr. Robert Austin from 1966 through 1981 on a Raub silt loam (Aquic Arguidoll) at the Purdue University Agronomy Farm. Raub developed on Wisconsin till, is poorly-drained with 0-1% slope, and has an organic matter content of about 3.5%. A number of full-season corn (Zea mays L.) hybrids were planted in 71-cm rows at a population density of about 54,560 plants per hectare throughout the experimental period. Each experimental plot was six rows wide and 21 m long. The specific corn hybrids used and the dates of planting for each year are shown in Table 1.

Nitrogen rates and application methods were replicated three times. The source of fertilizer N was ammonium nitrate in the early years and urea in the later years, except for the fall application of anhydrous ammonia, knifed in between the rows after harvesting. The application methods and rates of fertilizer N used in the experiment are shown in Table 2. The spring broadcast treatments were applied before planting. Check plots, which received no nitrogen, were also included each year. Corn grain yields for each experimental plot were determined from a 15-m length of the two center rows and expressed in kg/ha at 15.5% moisture. Since it took at least the first four years of the field trials for the corn yields to reach equilibrium levels on the low fertility treatments (0, 67 Kg N/ha), the 1966-1969 (SX-29) yields were not used in this paper.

Weather data necessary to calculate the components in ECG on a daily basis—solar radiation (SR), maximum and mimimum air temperatures used to compute FT, precipitation, and pan evaporation—were available from the

TABLE 1. *Corn hybrid, year planted, and date of planting in the long-term fertility management experiment, Field 54, Purdue University Agronomy Farm.*

CORN HYBRID	YEAR	PLANTING DATE
P-A-G SX29	1966	26 May
	1967	19
	1968	7
	1969	23
Pioneer 3369A	1970	19
	1971	20 Apr
	1972	18 May
	1973	14
	1974	6
	1975	8
	1976	23 Apr
AGRI 777	1977	11 May
Beck 65X	1978	31
	1980	6
	1981	1 Jun
Pioneer 3183	1979	10 May

TABLE 2. *Application method and Nitrogen source and rates of fertility used in the long-term fertility management experiment, Field 54, Raub silt loam, 1966-1981, Purdue University Agronomy Farm.*

APPLICATION METHOD AND N SOURCE	N FERTILITY RATE (kg/ha)
Fall Anhydrous Ammonia (1966-1978)	67,134,202
Fall Broadcast (ammonium nitrate or urea)	67,134
Spring Broadcast (ammonium nitrate or urea)	67,134,202
Spring Broadcast on even years (ammonium nitrate or urea)	134,269,403

cooperative National Weather Service-Purdue University weather station, located about 700 m south of the fertility experiment. The soil moisture model for poorly drained soils described by Dale et al. (1982) was used to obtain the daily estimates of ET/PET required in (1), assuming the soil moisture characteristics for Raub silt loam approximated those for Chalmers silt loam. Leaf area measurements were not taken in the long term fertility experiment, and it was necessary to estimate LAI to obtain the FLAI term in ECG. This was done with a model modified from that described by Dale, Coelho and Gallo (1980) for corn on high fertility soils. The algorithm used to predict the maximum leaf area per plant (LAMAX) was changed from a simple linear function of plant population density (P) to a function of P and its interaction with nitrogen rate (N): LAMAX = 0.834 -0.00578 P + 0.0000157 N X P. This function was developed with leaf area measurements made on these same fertility plots in 1978 and 1979 by Walburg et al. (1982) and in 1982 by J. P. McGarrahan and T. K. Flesch of the Agronomy Department, Purdue University.

Multiple regression models used in this study were of the general form:

$$Y = \beta_0 + \Sigma\beta_i X_i + \epsilon, \qquad (2)$$

where Y is the experimental corn grain yield for a given treatment-year β_0 and β_i are the regression coefficientes, X_i the independent variables, and ϵ the random error. The independent variables considered in this study were nitrogen rate (N), N^2, ΣECG, and the interaction, N X ΣECG, where Σ ECG is the sum of daily values (1) from ΣFT = 13 to 50 days after silking. The regression coefficients in (2) were fitted with computer programs from the Statistical Package for the Social Sciences (SPSS15) at the Purdue University Computing Center.

Results and Discussion

The average yields for all corn hybrids for the indicated N rate within each of the application methods, 1970-1981, are shown in Figure 1. The weather, however, causes year-to-year variability from these average yield responses to N (Broadbent, 1980). For example, for Pioneer Hybrid 3369A 28 sets of observations (7 years, 4 nitrogen treatments) were available to examine the N X ΣECG interactions for both the spring broadcast and fall anhydrous applications. A scatter diagram of Pioneer 3369A corn yield on ΣECG for each of the years and N levels is shown in Figure 2. At the 0 and 67 kg N/ha levels for the spring broadcast application there is little response to "better weather" as identified by greater ΣECG. At the 134 and 202 kg N/ha levels, however, there is a definite trend toward increasing yields with increasing ΣECG, as also shown on Figure 2 by the regressions fitted to the data and evaluated at 0, 67, 134, and 202 kg/N/ha.

Of the regression models examined by Mach (1982) the model $Y = b_0 + b_1N + b_2N^2 + b_3N \times \Sigma ECG$ consistently gave greater coefficients of determination (R^2) and F values, as well as the most consistent regression patterns. All regression coefficients and models were significant at the $\alpha = 0.05$ level. Because the linear ΣECG term is included only in the interaction term, the predicted slope of Y on ΣECG for N = 0 is necessarily 0. The quadratic form of the yield response to N can be seen in the greater yield increase from 0 to 67 kg N/ha from 67 to 134 and 134 to 202.

The increasing yield response to ΣECG at higher N is also evident in Figure 3 for the fall anhydrous ammonia application. The fitting of the quadratic model with the observational data, however, causes the predicted yields for the N = 202 level to be below those for the 134 level, except at greater ΣECG. In fact, the regression model shown in figure 3 provides a maximum predicted corn yield at 162 kg N/ha with ΣECG = 38 and at 125 kg N/ha with ΣECG = 28.

The scatter diagram of corn yields on ΣECG for the indicated year and N rate for the fall broadcast treatment is shown in Figure 4. There seems to be little yield response to ΣECG at any N level. The yield increases were attributed to increases in nitrogen rate. Some environmental or biotic factor was more influential in determining yield than the ΣECG variable, especially in 1973, which had the lowest yield and highest ΣECG. The mean yield level for N = 134 in Figure 4 is below that

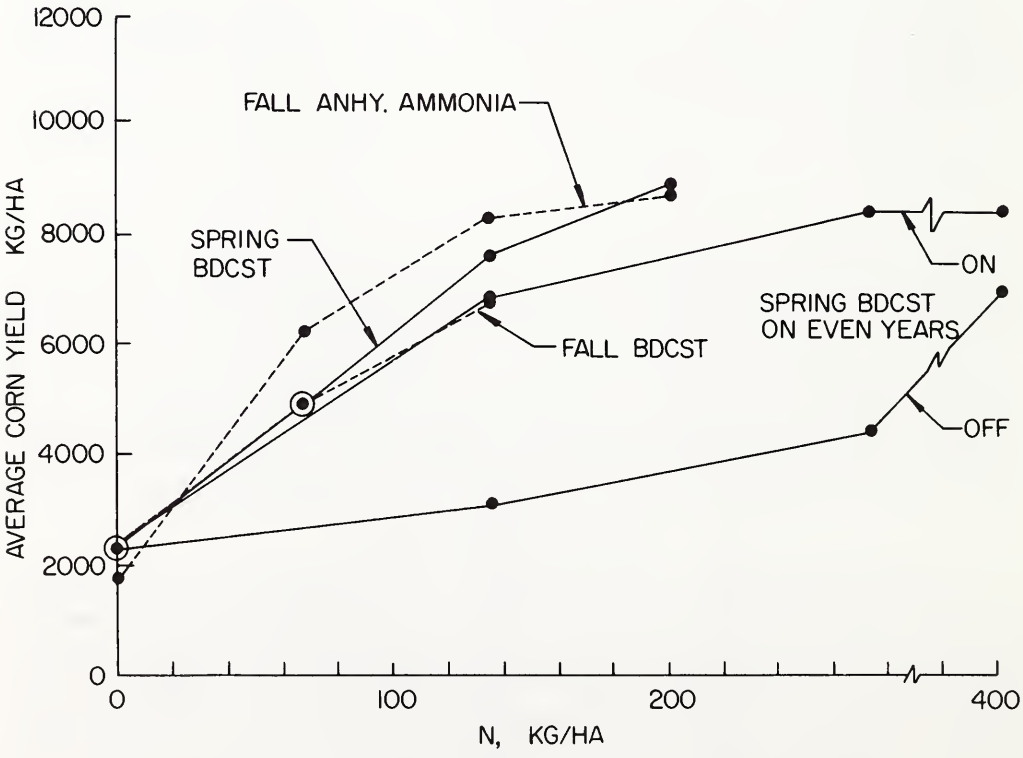

FIGURE 1. *Average response of corn yield to nitrogen rate (N) for indicated application method, 1970-1981, except fall anhydrous ammonia, 1970-1978, Raub silt loam, West Lafayette, Indiana. For the spring broadcast every other year, "ON" is the average yield for the years in which the fertilizer was applied, "OFF" for the years in which it was not applied.*

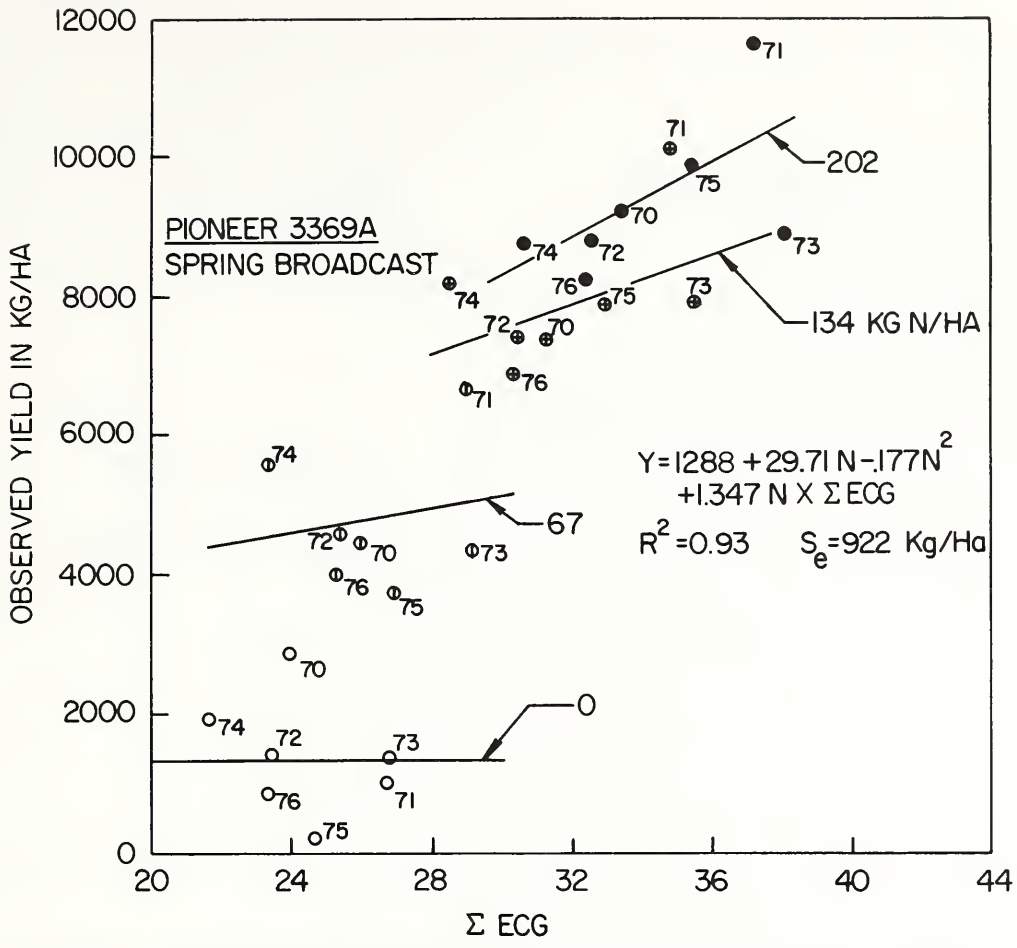

FIGURE 2. *Scatter diagram and regression fitting of corn yield on the energy-crop growth index (ΣECG) and indicated nitrogen rate (N), N applied by broadcasting ammonium nitrate or urea in the spring for the indicated year, Pioneer hybrid 3369A on Raub silt loam, West Lafayette, Indiana. The plotted numbers and symbols provide the year and N level for the experimental yield, e.g. open circles are O N, solid circles 202 kg N/ha.*

in Figures 2 and 3, suggesting that considerable nitrogen had been lost between the fall broadcast application and the period of vegetative corn growth the following spring (Stevenson and Baldwin, 1969). Welch et al. (1971) reported that excess water creates anaerobic conditions favorable for denitrification and that leaching also occurs when water percolates through the soil profile. Volatilization and leaching losses of the surface ammonium nitrate of urea can be affected by different weather conditions anytime from the nitrogen application to the beginning date for summing ECG, about 39 to 45 days before silking. Thus, the experimental N levels may not be as representative of the nutrient availability for the fall broadcast application as those for the fall anhydrous and spring broadcast applications.

Although there may be corn hybrid X nitrogen rate X weather interactions, the yields from all corn hybrids were pooled from 1970 within each of two application methods, fall anhydrous ammonia and spring broadcast. The regression model was fitted to the observed ΣECG, N, and corn yield data for each of these

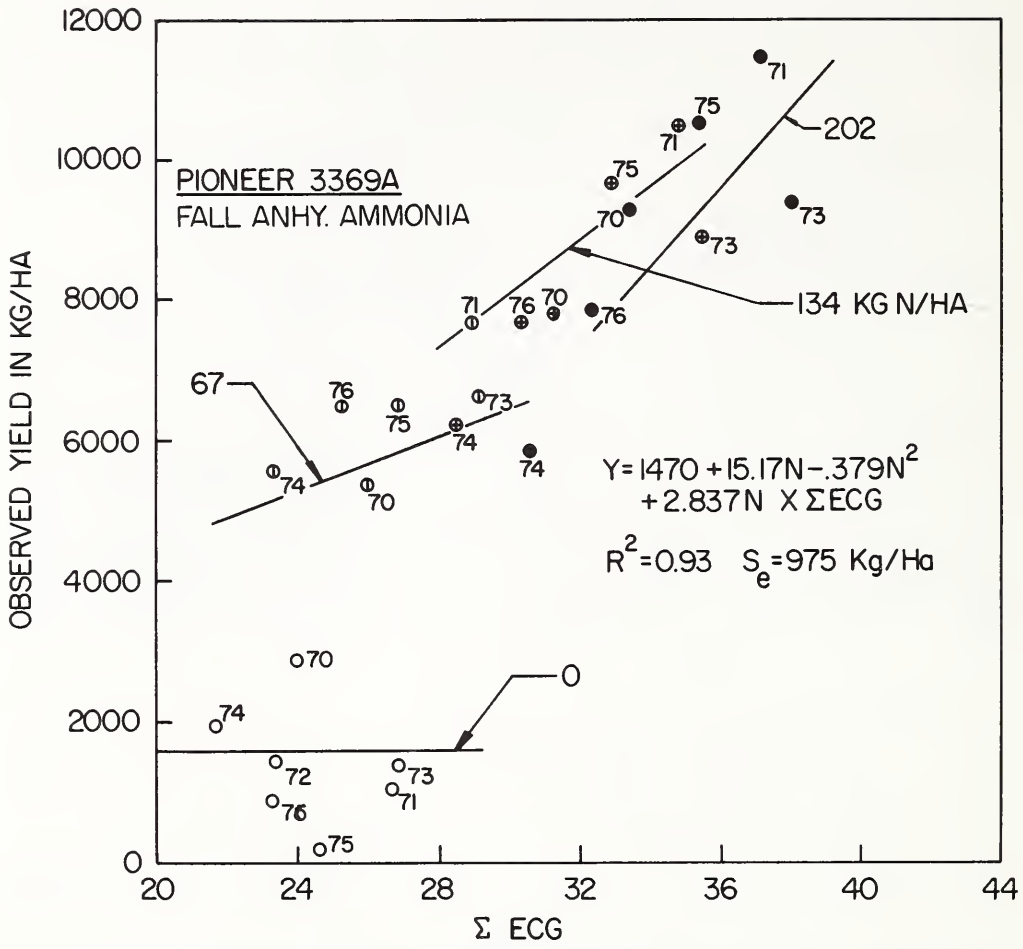

FIGURE 3. *Scatter diagram and regression fitting of corn yield on the energy-crop growth index (ΣECG) and indicated nitrogen rate (N), N applied by knifing in anhydrous ammonia the previous fall for indicated year, Pioneer hybrid 3369A on Raub silt loam, West Lafayette, IN. Plotted years and symbols same as Figure 2.*

management systems. In Figure 5 the observed yields were plotted against those estimated (predicted) for the fall anhydrous application, 1970-1978. The R^2 was 0.91, with the standard error 1010 kg/ha. Because there is no linear ΣECG term in the model, all of the predicted yields at N = 0 are at b_0.

The same regression model for the fall anhydrous application was used to predict yield for the indicated N at two levels of ΣECG, 28 and 38, "poor and good weather" years, respectively, as shown in Figure 6. These two curves generally bracket the overall average yields, reproduced from Figure 1 as the solid circles. The quadratic form of the model, previously discussed with the Pioneer 3369A results shown in Figure 3, indicates that corn yields predicted for any N with ΣECG = 38 were above those with ΣECG = 28, the difference increasing to a maximum of about 3800 kg/ha at the highest N level (202 kg N/ha). The yield response curve for ΣECG = 28 maximized at lower N than that for ΣECG = 38.

The regression model for the pooled corn hybrids for the spring broadcast application, 1970-1981, are also shown in Figure 6, evaluated for ΣECG = 28 and 38. Both curves are below those for the fall anhydrous application except at higher N and lower ΣECG values. The spring broadcast model does not reach a max-

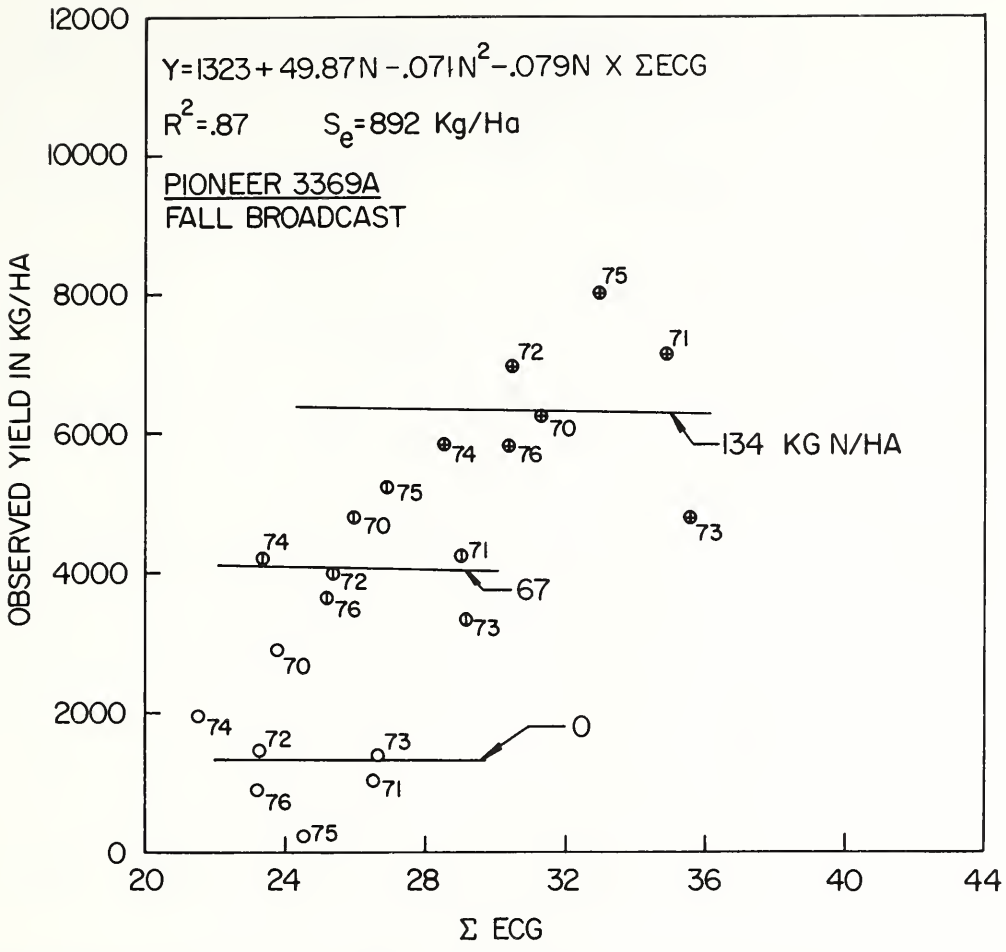

FIGURE 4. *Scatter diagram and regression fitting of corn yield to the energy-crop growth index (ΣECG) and indicated nitrogen rate (N), N applied by broadcasting ammonium nitrate or urea in the previous fall for the indicated year, Pioneer hybrid 3369A on Raub silt loan, West Lafayette, Indiana. Plotted years and symbols same as Figure 2.*

imum yield within the experimental N rates. The spring broadcast curves show less difference in yield response to N under low and high ΣECG than do the fall anhydrous curves.

Conclusion and Recommendation

A methodology for identifying weather influences on corn yield as a single energy-crop growth index, ΣECG, has been demonstrated and used to interpret corn yields from long-term fertility trials. Greater yield responses to increased levels of nitrogen fertilization occurred with higher than with lower ΣECG. Many improvements in the methodology could be made. First, some additional environmental measure of occurrences of warm weather, heavy rains, and other conditions known to be conducive to nitrogen losses from the time of fertilizer application through the vegetative growth period should be considered, especially when comparing fall with spring broadcast applications. Second, in this study corn growth was assumed to be equally responsive to ECG throughout the period

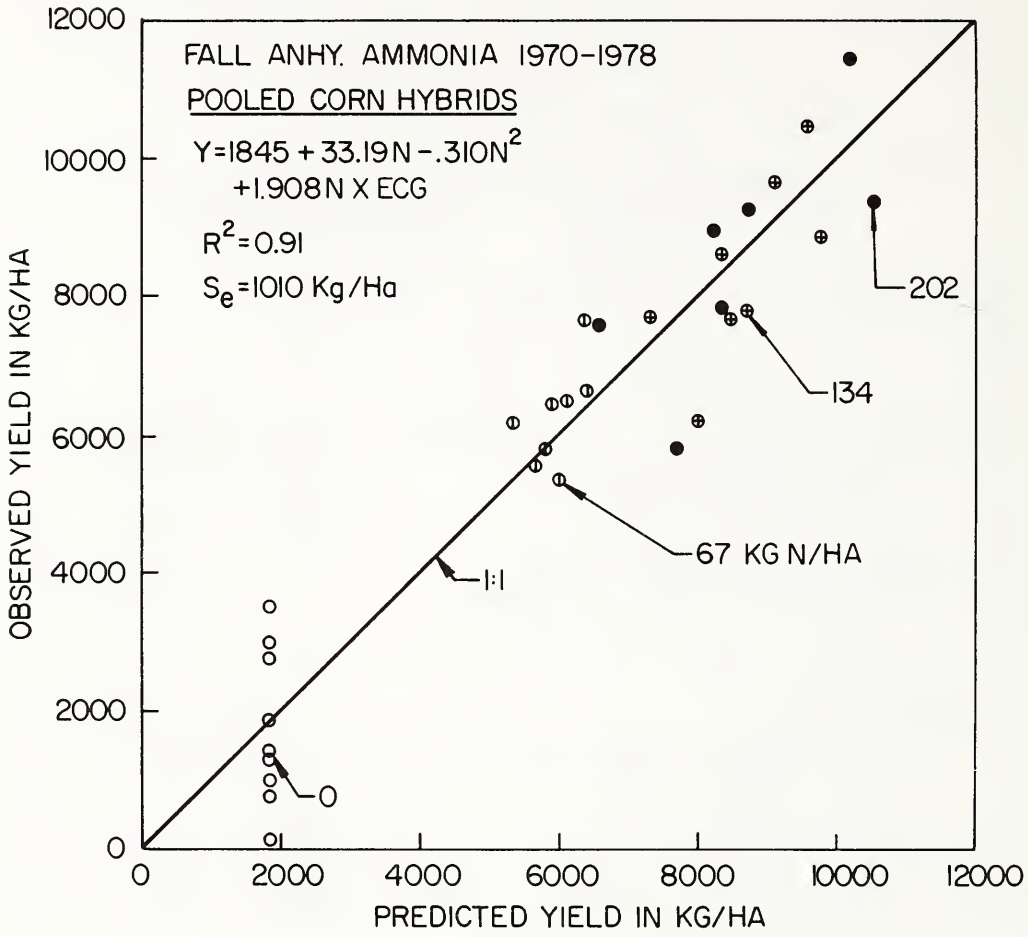

FIGURE 5. *Observed corn yield on that predicted with indicated regression equation and nitrogen rate (N), N applied by knifing in anhydrous ammonia the previous fall, for all hybrids used, 1970-1978, Raub silt loam, West Lafayette, Indiana.*

from ΣFT = 13 (39-45 days before silking) to 50 days past silking. Coelho and Dale (1980) showed this is unlikely. Weighting the components in ECG differently, such as weighting temperature heavier than solar radiation during the vegetative growth period and solar radiation heavier during the grain filling period, should be considered. Third, errors in measuring or estimating any of the ECG components for the field trials are multiplied in the ECG product variable. Finally, the ET/PET factor in ECG only provides "positive" plant moisture stress. "Negative stress", or too much water, may also be a significant factor in the early spring and should be examined.

Notwithstanding these self-critical statements, we suggest that the energy-crop growth variable may be useful in interpreting the effect of weather on corn yield response to nitrogen application in a set of long term fertility trials. The use of such a weather variable provides a means for pooling experimental data over years and locations to increase the range in the weather, crop variety, soil, nitrogen, and yield data, thereby increasing the inference space of the research results and expanding the soil-climate areas in which the results are pertinent.

Acknowledgments

This work was partially sponsored by the National Science Foundation, At-

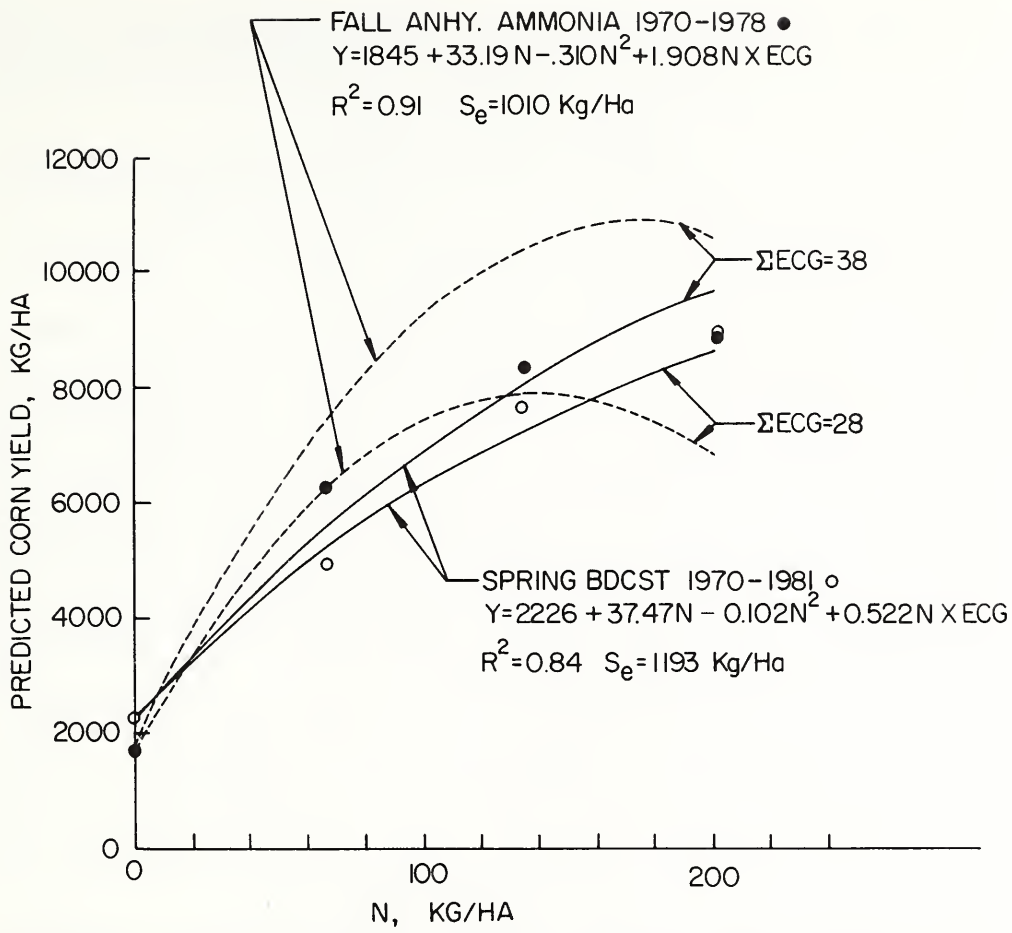

FIGURE 6. *Predicted corn yields on nitrogen rate (N) for two levels of the energy crop growth index, ΣECG = 38 ("good weather") and ΣECG = 28 ("poor weather"), and two application methods, fall anhydrous ammonia (1970-1978) and spring broadcast of ammonium nitrate of urea (1970-1981) for all hybrids used, Raub silt loam, West Lafayette, Indiana. The average yields for the indicated N rate have been reproduced from Figure 1, solid circles denoting fall anhydrous and open circles spring broadcast.*

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The Effect of Raindrop Impact and Slope Length on Sediment Concentration of Runoff

J. V. Mannering and W. C. Moldenhauer
Department of Agronomy and USDA-ARS National Soil Erosion Laboratory
Purdue University, West Lafayette, Indiana 47907

Introduction

The importance of raindrop impact on soil detachment has been recognized for many years (1, 2, 8). The importance of slope length and its influence on the total erosion process has also been recognized for some time (8). There is no question that high sediment concentration in runoff is associated with excessive erosion that leads to reduced soil productivity (4). In more recent studies (3), sediment bound phosphorus (P) has been shown to be the major form of phosphorus transported from agricultural watersheds into Lake Erie. A recent final report of the Corps of Engineers' "Lake Erie Wastewater Management Study" estimated that 66% of nonpoint P loadings originate from agricultural land (7). Nelson and co-workers in the Black Creek Study (3) showed that 93% of the total P delivered to the Maumee River was sediment-bound P. Reduction of sediment concentration of runoff from agricultural watersheds is clearly needed if water quality is to be significantly improved. In the study reported in this paper effect of raindrop impact and surface runoff in detaching and transporting soil material was determined. This is important in order to develop land treatment measures that will effectively reduce sediment yields from agricultural watersheds.

Materials and Methods

This study was a part of the EPA-supported Black Creek Watershed Sedimentation Study located in Allen County, Indiana (3).

ARS-Purdue rainfall simulator (6) tests were conducted at four locations during the months of July and August, 1974. These four locations consisted of four distinct soils, identified in Table 1, which were representative of over 80% of the major soil capability units contained in the 4800 ha Black Creek Study Area.

All test locations, which were in rowcrops the prior year, were moldboard plowed in early spring and disked twice immediately prior to conducting tests. The final disking was performed up and down the dominant slope. Metal borders were installed to delineate runoff plots 1.9 m wide and 10.7 m long at all locations. The simulated rain used in the tests closely approximated the drop-size distribution of natural rain and approached 80% of the kinetic energy of natural rain. Inflow was also simulated by introducing water at the upper end of the plots using two rates, one and two times the equilibrium runoff rate induced by rainfall. The four tests were all run sequentially on the same plot at each loca-

Table 1. *Identification of Test Locations*

Soil Phase	Classification	Percent Slope
Haskins loam	Aeric Ochraqualf; fine-loamy, mixed mesic	0.2
Nappanee clay loam	Aeric Ochraqualf; fine, illitic, mesic	0.7
Hoytville silty clay	Mollic Ochraqualf; fine, illitic, mesic	0.5
Morley clay loam	Typic Hapludalf; fine, illitic, mesic	5.1

tion. The first test was separated by the second, third and fourth tests by approximately 24 hours.

The test sequence was as follows:

First Test (Rain vs. Inflow Rate 1)—Simulated rain was applied for 45 minutes at 6.35 $cmhr^{-1}$, after which time the rain was turned off and inflow was introduced at the upper end of the plot for 15 minutes at approximately the same runoff rate as was occurring just before rainfall was cut off. This allowed a direct comparison of rain-induced runoff and inflow-induced runoff without rain on the sediment concentration of the runoff. At all four locations, the runoff from rainfall alone had reached equilibrium several minutes prior to the end of the 45 minute rainfall period. The inflow at the upper end of the plot was delivered by means of a gated aluminum pipe.

Second Test (Rain + Inflow Rate 1)—Thirty minutes of rainfall was applied at 6.35 $chmr^{-1}$ and inflow was simultaneously introduced at the upper end of the plot to produce twice the runoff achieved at the steady-state condition which would have occurred from rainfall-induced runoff alone. Once rainfall-induced equilibrium runoff was achieved, the rate remained essentially the same because of the somewhat stable surface seal produced. This second test enabled us to answer two additional questions. First, what effect does increased runoff rate (thus increased depth of water on the plot surface) have on the sediment concentration of runoff? Second, what effect does slope length have on the sediment concentration? It is assumed that doubling the runoff rate in essence doubles the effective plot length.

Third Test (Inflow Rate 1 vs Inflow Rate 2)—Inflow Rate 1 was applied for 15 minutes, followed by Inflow Rate 2 for another 15 minutes. The effect of doubling the runoff rate on sediment concentration could now be compared without considering the effects of rainfall energy. Inflow Rate 2 essentially doubled the effective plot length.

Fourth Test (Rain + Inflow Rate 2)—Simulated rain was applied at 6.35 $cmhr^{-1}$ for fifteen minutes, while simultaneously adding the inflow required to produce three times the steady-state runoff rate from rainfall alone. In effect, the slope length was tripled. This test expanded the information gathered from the second test showing how an even greater depth of water on the plot surface would effect the sediment concentration of runoff.

Data collected from these studies included: a) runoff hydrographs from which runoff rates were determined and b) 1% aliquot samples of runoff taken approximately every five minutes from which percent sediment was determined. Only the segments of hydrographs where both runoff and sediment concentrations were either approaching or were at equilibrium were used.

Results and Discussion

The runoff rate and sediment concentration data for all four test locations are given in Table 2. In the first test approximately 75% of the 6.35 $cmhr^{-1}$ rainfall application was lost as runoff at the time sediment concentrations were measured at all four test locations. This was caused by the action of the raindrops in developing a slowly permeable surface seal. Once this surface seal was formed, it became the dominant factor restricting infiltration. Earlier simulated rainfall tests under conventional seedbed conditions showed that a relatively stable surface seal occurred on a broad range of soils within the first twenty minutes of rain application (5). It was assumed that the surface seal remained fairly stable through the rest of the test sequences.

TABLE 2. *The effect of raindrop impact and slope length on sediment concentration of runoff*

Soil Phase	Test Sequence	Runoff Rate[1] $cmhr^{-1}$	Sediment Concentrations %
Haskins loam	1st Test - Rain	4.8	1.18
	Inflow Rate 1	4.8	0.18
	2nd Test - Rain + Inflow Rate 1	9.6	0.91
	3rd Test - Inflow Rate 1	4.8	0.40
	Inflow Rate 2	9.6	0.48
	4th Test - Rain + Inflow Rate 2	—	—
Nappanee clay loam	1st Test - Rain	4.3	0.98
	Inflow Rate 1	4.3	0.08
	2nd Test - Rain + Inflow Rate 1	8.6	0.54
	3rd Test - Inflow Rate 1	4.3	0.02
	Inflow Rate 2	8.6	0.03
	4th Test - Rain + Inflow Rate 2	12.9	0.30
Hoytville silty clay	1st Test - Rain	5.3	1.37
	Inflow Rate 1	5.3	0.14
	2nd Test - Rain + Inflow Rate 1	10.6	0.49
	3rd Test - Inflow Rate 1	5.3	0.09
	Inflow Rate 2	10.6	0.09
	4th Test - Rain + Inflow Rate 2	15.9	0.29
Morley clay loam	1st Test - Rain	5.3	3.81
	Inflow Rate 1	5.3	0.42
	2nd Test - Rain + Inflow Rate 1	10.6	2.43
	3rd Test - Inflow Rate 1	5.3	0.34
	Inflow Rate 2	10.6	0.41
	4th Test - Rain + Inflow Rate 2	15.9	2.92

[1]Runoff rates are the average runoff rate measured over a 30 minute rainfall period after a surface seal was well established and runoff had reached an equilibrium condition.

Haskins loam (0.2% slope)

Raindrop energy was a major force affecting sediment concentration for Haskins loam since inflow-induced runoff in the first test was only 15% of that for rain-induced runoff. This conclusion was further supported by the results of the second test, where the re-introduction of rainfall in conjunction with inflow dramatically increased sediment concentration compared to inflow alone. This value (.91%) was slightly lower than the value (1.18%) for rain alone in the first test which may suggest that the increased runoff from the inflow cushioned raindrop impact slightly and thus, its detachment capability, a conclusion that could have some merit on nearly level topography.

The third test sequence showed that doubling inflow alone increased sediment concentration only slightly (.40 to .48%). This suggests that increasing slope length had only a slight effect on sediment concentration at this site. Insufficient water was available to complete the fourth test on the Haskins soil.

Nappanee clay loam (0.7% slope)

The case for raindrop energy being the principal force affecting sediment concentration is even stronger for the Nappanee soil than for the Haskins soil. Results of the first test show that the value for inflow-induced runoff to be only 8% of that for rain-induced runoff. Again a dramatic increase in sediment concentration occurred when rain was included with inflow in the second test sequence.

The cushioning effect of the increased runoff depth from the inflow, as in the case of the Haskins soil was responsible for the lower sediment concentration with rain plus inflow compared to rain alone (0.54% vs 0.98%) since this soil was also situated on essentially level topography.

The third test further strengthens the evidence that inflow-induced runoff has little effect on sediment concentration of nearly level soils at least for the flow rates that were used. Values for both the single and double rates of inflow were essentially the same and were extremely low. Evidently, runoff velocity on this nearly level soil was not sufficient to detach or transport significant amounts of sediment.

In the fourth test, where rain was added to the double inflow rate, the sediment concentration was only about 30% of that resulting from rain alone. This latter comparison supports the concept that increased runoff depth cushions the impact of raindrops. Also, tripling the slope length as simulated in the fourth test appeared to have limited influence on sediment concentration from this nearly level soil.

Hoytville silty clay (0.5% slope)

This soil responded similarly to the Nappanee and Haskins soils. Results from the first test showed the sediment concentration from inflow-induced runoff to be only 10% of that for rain-induced runoff. Also results from the second test showed that the addition of rain to inflow increased sediment concentration, but the value was appreciably lower than with rain alone. In the third test, no difference in the sediment concentrations was noted when inflow amount was doubled. Runoff from nearly level land has extremely limited soil detachment and transport capacity. In the fourth test, sediment concentration from the double inflow rate plus rain was only 21% of that from rain alone. Again the increased flow depth reduced the detachment energy of the rainfall.

Morley clay loam (5.1% slope)

The Morley soil site was the only test location that had appreciable slope. This slope markedly influenced the sediment concentration. For example, in the first test with rain alone, the sediment concentration was approximately three times that of the other nearly level sites. This result was generally consistent in all of the tests where rain was included as part of the treatment. Evidently, the greater slope increased the runoff velocity and its sediment transport capacity sufficiently to remove a much greater proportion of soil detached by raindrop impact. Another factor that could be involved is the absence of surface ponding (which occurred on the nearly level soils, thus providing protection from raindrop impact) on the more sloping Morley site.

Although runoff velocity obviously increased at this site because of steeper slope, it still lacked sufficient energy to detach soil particles in amounts which would result in high sediment concentrations. Raindrop impact was needed, even on a 5% slope, to detach sufficient soil to create a serious problem. Results from the first test showed sediment concentration from inflow-induced runoff to be only 11% of that for rain-induced runoff while the inclusion of rain with inflow in the second test dramatically increased the sediment concentration. It was still only 64% of the value for rain alone. This result suggests that slope length has some impact on sediment concentration, but that raindrop impact still remains the major force for these rainfall and runoff rates. However, when the slope length is doubled without rain, sediment concentration is affected very little. Results

from the fourth test showed that the additional flow volume increased the sediment concentration when compared with the second test. This increase did not occur on the nearly level sites with the Nappanee and Hoytville soils.

Summary

The results of the simulated rainfall and inflow studies conducted on four soils in the Black Creek Watershed in Allen County, Indiana, show rainfall energy to be the major factor affecting sediment concentration of runoff from agricultural soils in a seedbed condition. Sediment concentration of rainfall-induced runoff ranged from 7 to 12 times that of inflow-induced runoff for all the soils. In the study, doubling or tripling the selected inflow to simulate increased slope length had little influence on sediment concentration. However, slope steepness did influence sediment concentration. On the test site having the 5.1% slope, sediment concentrations approximately three times those from the other three nearly level test sites occurred whether runoff was rainfall-induced, inflow induced, or a combination of both. These results demonstrate again the necessity of protecting the soil surface from raindrop impact in order to effectively reduce sediment concentrations of runoff which will, in turn, maintain soil productivity and enhance water quality.

Acknowledgment

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ZOOLOGY

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Saint Mary's College, Notre Dame, Indiana 46556

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ABSTRACTS

Annual Reproductive Cycle of the Female Mosquitofish (Gambusia affinis) in Taiwan. YUN-CHIA CHOU and PAUL S. ALEXANDER, Department of Biology, Tunghai University, Taichung, Taiwan 400, Republic of China.——From January, 1979 to June, 1980 monthly samples of *Gambusia* were netted from spring-fed irrigation ditches in suburban Taichung City. Pregnant fish were found in every month except November and December and comprised about half of the 847 females with total body lengths of 24-61 mm. The rate of pregnancy, based on observation of "eyed" embryos, ranged from 14.0% in January and October to 68.0% in March and June. There was a tendency for the minimum size of pregnant fish to decrease from January (35 mm) to August (21 mm). Only one fish shorter than 24 mm was found to be pregnant (21 mm in August). Winter-born fish began to mature in April at minimum body length of 28 mm. The high mortality of larger fish beginning in May presumably followed production of the third brood.

These data taken at about N latitude 24° are compared with those reported for the same species from the United States (N latitude 30°-41°) and Japan (N latitude 36°). The influences of photoperiod and temperature on reproduction of *Gambusia affinis* are discussed.

The Possible Role of Endurance Training in Diabetic Microangiopathy. LARRY GANION, Ball State University, Muncie, Indiana 47306.——To examine the role endurance training may play in the pathogenesis of microangiopathy in capillaries of diabetic muscle, samples were obtained by needle biopsy from the gastrocnemius of human subjects at the commencement and completion of a ten week training program and prepared for electron microscopy. The participants, four non-diabetics and four diabetics, ran five days per week at a stress level of 75.0% of their maximum work load. The muscle samples were immersed in 3.0% glutaraldehyde for four hours, rinsed in 0.1M Sorenson's phosphate buffer (pH 7.2), postfixed for one hour in 1.0% osmium tetroxide, and embedded in Epon 812. Ultrathin sections were cut on a Porter-Blum MT-2B ultra-microtome, stained with uranyl acetate and lead citrate, and viewed with an RCA EMU-3C electron microscope. Microangiopathy is a vascular disease characterized by the thickening of the capillary basement membrane (CBMT). The CBMT was analyzed via measuring the width of the capillary basement membrane in electron micrographs prepared from muscle samples. The results of this study confirm those of previous studies, namely that the capillary basement membrane of diabetics is thicker than the non-diabetic. The mean CBMT of the diabetic pretrained group and non-diabetic pretrained group respectively were 2598Å and 1830Å. Measurements of the capillary basement membranes revealed that the CBMT of the trained diabetic muscle was less than CBMT of the untrained muscle. The untrained group displayed a mean CBMT of 2598Å and the trained group had a mean CBMT of

2122 Å. These preliminary data suggest that endurance training may favorably alter the pathology of microangiopathy in diabetics.

Effect of Amoscanate on the Tegument of Schistosoma mekongi. WILMAR B. JANSMA, Department of Life Sciences, Indiana State University, Terre Haute, Indiana 47809.—— Scanning electron microscopy was utilized to study tegumental alterations of *Schistosoma mekongi* resulting from exposure to the anti-schistosomal drug Amoscanate. Female mice (CD-1, Charles River Laboratories, Wilmington, MA) harboring adult *S. mekongi* were treated with a curative oral dose of 7 mg/kg b.w. Amoscanate (4-isothiocyanate-4′ nitrodiphenylamine, CGP 4540). Mice were sacrificed by cervical dislocation and worms recovered from host mesenteric veins, hepatic portal vessels and/or liver sinuses at 1, 3, 6, 12, 24 hours and 4, 7, 10, 14, 21, and 24 days post-treatment. Worms recovered were fixed and processed for scanning electron microscopy via critical point drying techniques.

Alteration of the tegument of male *S. mekongi* was observed as early as 1 hour after drug exposure. Folds of the tegument initially ballooned outward and progressive damage resulted in large flattened outpocketings as the tegument lost its overall integrity. After 24 hours, nearly all worms had shifted to the liver and widespread cratering of the surface and shrinkage of the tegument was observed. Worms recovered from the liver at later times (10, 14, and 21 days post-treatment) showed progressive surface damage and large tegumental regions were denuded exposing underlying tissue. Numerous host blood cells were observed attached to the male tegument. The entire surface of male worms recovered 24 days after treatment appeared as a sponge-like layer, being highly cratered and vacuolated and lacking any resemblance to the normal highly convoluted surface of *S. mekongi*.

Female *Schistosoma mekongi*, in contrast, were far less affected by the schistosomicidal drug Amoscanate. Only slight damage to the female surface was noticed after 4 days and only after 14 days was extensive tegumental damage observed on the surface of female worms. Tegumental alterations consisted of uniform raised protuberances over the entire female surface.

Evidence of tegumental damage caused by anti-schistosomal drugs may provide clues regarding the biochemical mode of action of these drugs and their cytocidal foci within these medically important parasites.

Further Information on the Specificity of the Male Cricket, Acheta domesticus (L.), Requirement for Vitamin E. JAMES R. LITTON, JR., Department of Biology, Saint Mary's College, Notre Dame, Indiana 46556.—— Vitamin E is required for spermatogenesis in the house cricket. While selenium and tocopherol quinone cannot substitute for α-tocopherol (vitamin E), the further specificity of this requirement for vitamin E had not been exhaustively investigated.

Antioxidants (ethoxyquin and menadione), selenium (with and without methionine), hexahydro coenzyme Q_4, tocopheronolactone, N-methyl-γ-tocopheramine, 5,5′-methylene bis (γ-tocopherol), 2,5,7,8-tetramethyl-2-(4′,8′-dimethylnonyl)-6-hydroxychromane, and a series of chromanol derivatives were compared with recent data for the tocol and tocotrienol series. All of these derivatives showed a varied pattern of substitution for natural *d*-γ-tocopherol. The newly evaluated compounds ranged in activity (*d*-γ-tocopherol = 100%) from 0-34.6% based on assay of egg viability and examination of the testes for the presence of motile sperm.

Information on the relative effects of these compounds on larval and adult

growth rates was also obtained. The specificity and pattern of response of crickets to these and previously tested compounds facilitates a comparison of cricket, rotifer and mammalian vitamin E requirement.

The Marine Interstitial and Planktonic Rotifer Fauna of Puerto Rico. JAMES R. LITTON, JR., Department of Biology, Saint Mary's College, Notre Dame, IN 46556.——Investigations of the fauna of coral reefs and surrounding waters have never carefully examined the microfauna, and specifically the rotifers, in a detailed manner. While rotifers are predominantly freshwater forms they can be regularly found in the interstitial and planktonic fauna of marine systems if particular care (mesh size of nets and detailed sample sorting techniques) is given to sample collection.

Five marine sites in Puerto Rico (southern shore coral reef, northern coral reef, Phosphorescent Bay, a mangrove cay, and northern shore coral sand) were sampled during daylight hours using a 125 mesh (No. 25) nylon plankton net. Wet sand samples were placed in the net, eluted with a known volume of seawater, and treated as plankton samples. All samples were examined live and then preserved in 10.0% formalin for later specific and quantiative observations.

The following rotifer genera were found in the samples: *Keratella*, *Notholca*, *Philodina*, *Collotheca*, *Lecane*, *Proales*, and *Lepadella*. The diversity, abundance, and salinity tolerance of these genera and species forms an interesting comparison with data from northern European and north American marine locations. While the rotifer component of the marine plankton and interstitial fauna may not be large it is probably always present.

The Effect of Single and Divided Dose Administration on the Efficacy of Cambendazole against Trichinella spiralis. R. O. MCCRACKEN, A. GARCIA, D. NIERSTE and D. DOCK, Department of Biology, Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana 46223.——Mice infected with adult *Trichinella spiralis* were used to compare the anthelmintic efficacy of cambendazole (CBZ, 2-[4′-thiazolyl]-5-*iso*propoxycarbonylamino-benzimidazole) given as a single oral dose or administered as a series of divided daily oral doses during the enteral phase of infection. Given as a single oral dose of 300 mg/kg 72 hr after exposure to infection, CBZ reduced the worm burden by 61.0% as determined at necropsy on day 7 postinoculation; as a single dose of 600 mg/kg it removed 88.0% of the adult worms. Twice daily oral doses of CBZ at 25 mg/kg for 3 consecutive days during the intestinal phase (i.e., a total dose of 150 mg/kg body weight) was equally effective against the drug resistant adults; this treatment regimen reduced the worm burden by 79.0%. The demonstrated role of the duration of exposure to the anthelmintic as a determinant of its efficacy and spectrum of action has important therapeutic implications for extending the range of life cycle stages of *Trichinella* against which CBZ is effective, and for achieving efficacy with low dosage rates.

Ontogenetic Lipid Metabolism of Abystoma tigrinum. ERIC D. MOULD and DAVID M. SEVER, Department of Biology, Saint Mary's College, Notre Dame, Indiana 46615.——Collections of *Ambystoma tigrinum* were made at weekly intervals from a pond in St. Joseph Co., Indiana from March 22, 1982 when egg masses were observed until August 16, 1982 when no salamanders remained in the pond. Lipid content of the collected specimens was determined by Soxhlet extraction and a randomly selected sample was saved for length measurements and stomach analysis. Lipid content of egg masses was 10.81% and was generally lower in subsequent larval collections, presumably due to yolk consumption. From late

May until collections were terminated, larva varied dramatically in size, two to three fold variation in snout-vent (SV) lengths were common. In latter stages of larval development, lipid content was directly correlated with S-V length. Ecological and evolutionary consequences of the diversity in larval length, lipid content and developmental stage are considered.

Differences in Feeding Patterns among Three Age Groups of Walleye (Stizostedion vitreum vitreum) in Western Lake Erie. ELLEN K. PIKITCH, Department of Biology, Indiana University, Bloomington, Indiana 47405.——Information concerning feeding patterns is often a critical component in the management of fish populations. Studies of feeding patterns are most useful when they take into account not only the species of prey consumed, but also the availability of various forage species in the environment. However, when different predator age groups possess differential capabilities for utilizing prey, the concept of forage availability becomes more complex.

To examine age-specific forage availability and utilization, walleye and their potential prey were collected over a seven month period in 1981 from western Lake Erie. Nearly 2,200 walleye were collected, of which only 65.0% contained food. It was found that the frequency of feeding decreased, whereas numbers and size of prey consumed increased with age. The walleye diet varied from month to month in response to changes in the forage community. Generally, the dominate species in the diet were those that were most abundant in the environment, but there were some notable exceptions. In addition, young-of-the-year (y-o-y), yearling and older walleye utilized different prey species during each month. These differences in prey selection can largely be explained by examining the size distribution of each prey species during each month, in relation to the average gape of each age group of walleye.

Of particular interest was the differential utilization of the gizzard shad (*Dorosoma cepedianum*). Gizzard shad grow much more rapidly in western Lake Erie than in most other lakes. A previous study of y-o-y and yearling walleye feeding habits in Lake Erie did not find gizzard shad to be a significant component of the diet. The combination of high growth rates and lack of evidence to the contrary led to the belief that gizzard shad were an insignificant source of food for western Lake Erie walleye. In contrast, this study shows that when gizzard shad are abundant, they are an important source of food for all age groups of walleye, but at different times of the year. From July through October, gizzard shad consecutively grew into the preferred size ranges of y-o-y, then yearling, and finally older walleye. Consequently, the percent of the walleye diet composed of gizzard shad peaked in August, September and October for y-o-y, yearling and age II+ walleye, respectively. Gizzard shad comprised over 65.0% of the diet of each age class during its respective peak month.

Miscellaneous Notes on *Ambystoma texanum* in Vigo County, Indiana

Barbara A. Brown, Wynn W. Cudmore,
and
John O. Whitaker, Jr.
Department of Life Sciences, Indiana State University,
Terre Haute, Indiana 47809

Introduction

Ambystoma texanum, the small-mouth salamander, was studied extensively between 1974 and 1980 in Vigo County, Indiana. These salamanders congregate in temporary breeding ponds in early spring (Cook, 1964; Bailey, 1943). It is believed that *Ambystoma* spp. migrate above ground at night into the ponds (Taylor, 1972; Shoop, 1965), and that moisture and temperature together act as stimuli to migration (Blanchard, 1930; Wright and Allen, 1909). The purpose of this study was to gain an understanding of the movements of the small-mouth salamander during the breeding period and to determine the effects of temperature and precipitation upon migration.

Methods

Salamanders were collected at four sites in Vigo County, here referred to as Dave's Pond, Ambystoma Pond, Scaphiopus Pond and the Milwaukee Railroad embankment, in the early springs of 1974, 1976, 1978 and 1980. The first three ponds are in the uplands about 5-10 miles NE of Terre Haute. The railroad embankment is in the Wabash floodplain. Included in this study area is a breeding pond, but also, large numbers of salamanders live along the embankment itself, and numbers can often be caught under the old ties strewn along the tracks. This gave us a rather unique opportunity to collect information on this species during the non-breeding season

The animals were collected in minnow traps, along drift fences and, in 1980, in a specialized can trap set in the banks of two ponds (Ambystoma Pond, Railroad embankment), and under the ties (RR tracks). The drift fences consisted of ¼″ mesh hardware cloth 8″ high and of various lengths. Sunken bottle traps were placed along their lengths. These fences were established at varying distances from the ponds. The specialized can traps consisted of a metal can (10.5 cm diameter, 17.5 cm height) with a wire screen at one end and a wire screen funnel (5.5-6.0 cm in length) at the other. The funnel, with a 2 cm diameter opening at one end, was placed in tunnel openings along the banks of the ponds, at or near the water's edge. Traps were checked daily, sometimes more frequently, from the time they were set out until after the salamanders had left the ponds.

Some of the animals taken in traps were toe-clipped for identification and released in order to assess movements and duration of stay in the ponds. Population estimates (Peterson Estimate, Lincoln Index) were also calculated based on mark and recapture data.

Temperature and precipitation data were taken from climatological observations made by the Pfizer Company of Terre Haute in 1974 and from NOAA monthly climatological publications for Indiana in 1976, 1978, and 1980. Maximum and minimum temperatures were averaged for mean daily temperatures.

Results and Discussion

Earliest breeding reported for *Ambystoma texanum* is in December in Tennessee (Ashton, 1961). Generally small-mouth salamanders breed from March to April (Cook, 1964; Bailey, 1943; Smith, 1934). Results in this study show animals beginning to arrive in the breeding ponds in high numbers as early as 17 February 1976 and as late as 23 March 1978. It appears that the animals will not enter the ponds in Vigo County in January even if conditions are right (high temperatures, melting of ice). For example, in late January 1969 five days of 50 to 70°F temperatures occurred, and the ice completely melted from the ponds, but no salamanders were found in the ponds.

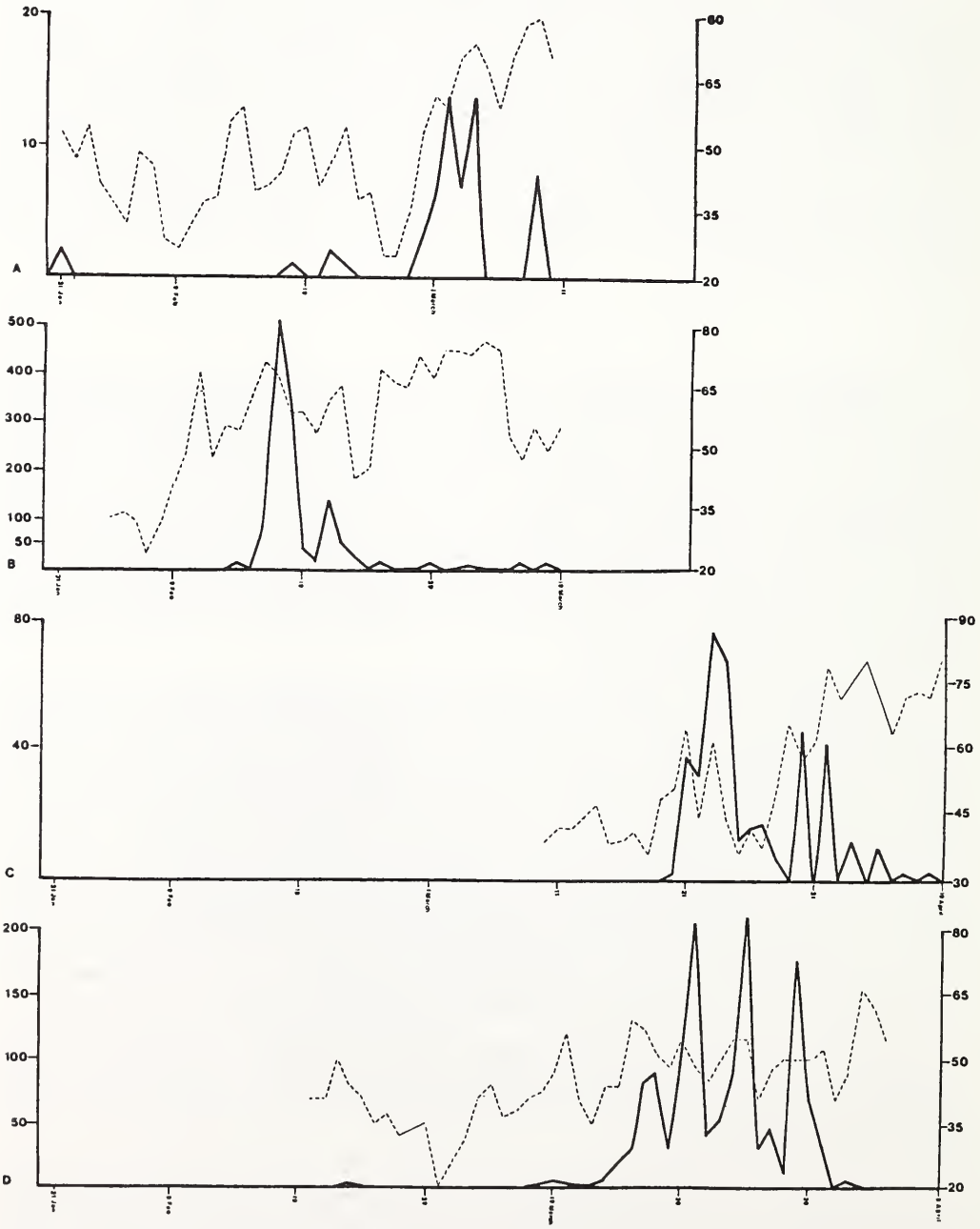

FIGURE 1: *Numbers of Ambystoma texanum Captured per Day (solid line) and Maximum Daily Temperatures (°F) for (A) 1974, (B) 1976, (C) 1978 and (D) 1980*

Temperature and precipitation are considered important as stimuli for migration into the breeding ponds. Smith (1934) reports animals breeding soon after the ponds were free of ice, and Bailey (1943) notes breeding associated with warm weather. Ramsey and Forseyth (1950) state that *A. texanum* in Texas breeds after heavy rains, while Bailey (1943) reports that although breeding may be associated with rains it is not considered a necessity. Minton (1972) states that two days or more of mild, rainy weather in winter stimulates migration to breeding ponds.

The influence of temperature on migration to the ponds seems obvious from our results (Figure 1). In 1974 and 1976 maximum daily temperatures were above 50°F on nearly all days when salamanders were captured. In 1978 and 1980 maximum temperatures were 40°F on all days but two in 1978 (25 and 27 March) when animals were taken. In all years the greatest number of animals per day was taken following at least four (1974) and up to 11 days (1980) of greater than 40°F maximum temperature. In 1974, 1976, and 1980 these peaks followed four (28 February, 1, 2 and 3 March; N = 7), five (13, 14, 15, 16, 17 February; N = 488) and three days (23, 24, 25 March; N = 210), respectively, of greater than 50°F maximum temperatures. In 1978 animals were beginning to peak on 20 March (65°F maximum) when temperatures dipped to 44°F and the numbers dropped. The next day temperatures rose to 62°F and the number of *Ambystoma texanum* increased accordingly (Figure 1C).

Precipitation did not produce a similar effect (Figure 2). There did not seem to be any relationship between precipitation and either peak numbers of animals taken in the ponds or the beninning of migration. In 1974 animals began to enter the breeding ponds on 18 February (N = 1). There was no precipitation on this day (Figure 2A). On 28 February animals again began to move into the ponds and again there was no precipitation, nor had there been any for two days prior to this. Numbers of animals peaked on 3 March (N = 14) and again on 5 March (N = 14). There was no precipitation on 3 March, but it rained 0.65 inches on 5 March.

In 1976 animals were first captured in the ponds on 13 February (N = 3) and increased over the next 4 days, to peak on 17 February (N = 488). There was no precipitation on 13 February; however, 1.05 inches of rain did fall on the seventeenth (Figure 2B).

Similar results were seen in 1978 and 1980. In 1978 migration began on 20 March with no precipitation (N = 2), and animal numbers peaked on 23 March with no precipitation (N = 78), and again on 30 March (N = 46), 0.05 inches of precipitation (Figure 2C). In 1980 animals began to enter the ponds on 9 March (N = 2), no precipitation, and peaked on 21 March (N = 205), .10 inches precipitation, and 25 March, no precipitation (N = 210) (Figure 2D). There did not seem to be any relationship between precipitation and movements of *A. texanum.*

Ambystoma spp. have been reported to migrate into breeding ponds at night (Hurlbert, 1969; Shoop, 1965). Similar, though limited, results were seen in this study. In 1974 at Dave's Pond between 3 March and 5 March, though traps were checked both in the morning and in the evening, all animals (N = 33) except 3 were captured during the night. In 1976, 1978 and 1980 traps were checked only once each day so no further data are available.

It was assumed that *Ambystoma texanum* travels above ground into the breeding ponds. However, results here seem to contradict this. In 1974 at Dave's Pond drift fences and minnow traps were set out by 24 January. The first salamander was taken in a seine on 31 January and 14 *A. texanum* eggs were seined on 14 February. It was not until 18 February that animals were captured at the fences. In 1980 minnow traps were set at Dave's Pond under ice. Two

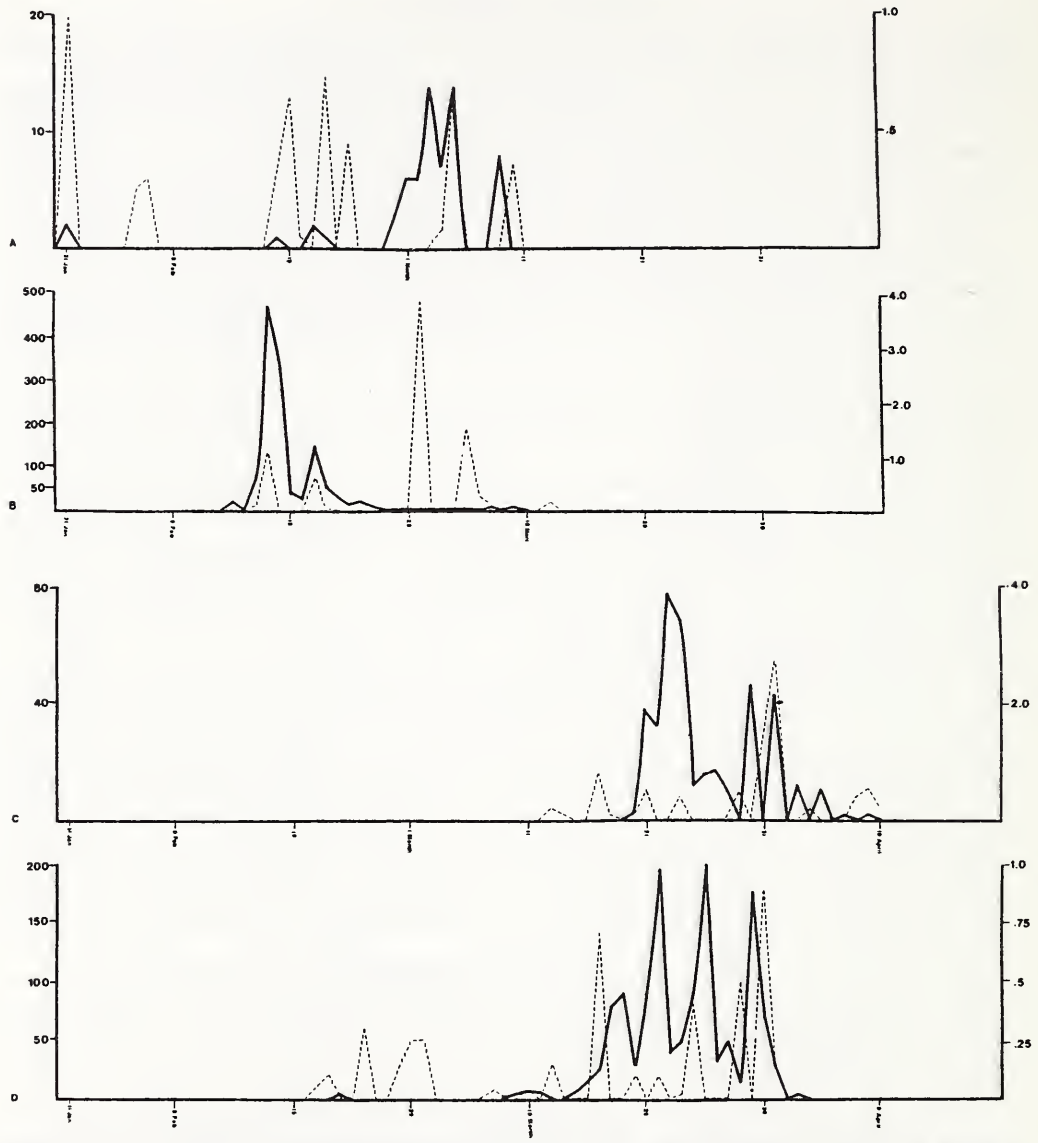

FIGURE 2: *Numbers of Ambystoma texanum per Day (solid line) and Daily Precipitation in inches.*

salamanders were captured on 23 February while the pond was still covered with ice. In that year specialized can traps were set in burrows in the banks of Ambystoma Pond and the Milwaukee railroad embankment pond. Ten *Ambystoma* were captured in these traps while none were taken at the fences. These results indicate that numerous small-mouth salamanders move underground through burrows into the ponds rather than above ground.

Animals were toe clipped and 1976 data were recorded on movements of the salamanders both in the breeding ponds and in a permanent colony found under railroad ties at the Milwaukee railroad embankment. At Ambystoma Pond (N = 22) animals moved an average of 12.7 meters and a maximum of 40 meters. At the Milwaukee railroad embankment (the permanent population) movements (N = 8) ranged from 0 to 170 meters $\bar{x}$ = 44.6 meters).

Mark and recapture data were also used in 1976, 1978, and 1980 to gather information concerning duration of stay in the breeding ponds. Data indicate that there is a rapid turnover of individuals, so the population estimates were not

TABLE 1: *Mark and Recapture Data Showing Duration of Stay in Breeding Ponds*

	Days since Marking																	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	Total
1978 (*N = 233)																		
Ambystoma ♂	26	23	18	12	3	5	4	19	11	3	12	1	1	0	0	0	1	139
Pond ♀	5	1	1	0	1	0	0	0	2	0	0	0	0	0	0	0	0	10
Total	31	24	19	12	4	5	4	19	13	3	12	1	1	0	0	0	1	149
1980 (N = 487)																		
Milwaukee ♂	3	2	2	2	4	0	3	1	0	3	0	2	0	0	0	0	0	23
Railroad																		
Pond ♀	0	0	0	0	0	0	1	0	0	0	0	1	1	0	0	0	0	2
Total	3	2	2	2	4	0	4	1	0	3	0	3	0	0	0	0	0	25
1980 (N = 120)																		
Ambystoma ♂	2	4	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	7
Pond ♀	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4
Total	6	4	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	11

*(N = Number of salamanders marked)

reliable. In 1976 on 16 February 17 individuals were marked and released. On 17 February of 109 animals captured 5 were recaptured. On 18 February there were 16 recaptures in 75 captures, 21 February 6 recaptures of 81 individuals, and on 22 February 6 recaptures of 27 individuals. Between 17 and 22 February 292 animals were captured with a total of 33 recaptures.

In 1978, two hundred thirty-three animals were marked and 147 were recaptured. In 1980 at Ambystoma Pond 120 individuals of *Ambystoma texanum* were marked in a total of 183 captures. Only 11 of these marked individuals were recaptured. That year at the temporary pond at the Milwaukee railroad embankment 1122 animals were captured, 487 were marked but only 24 recaptured. These low numbers of recaptures seem to indicate a rapid departure of the salamanders from the breeding ponds.

Males occurred in higher numbers in the breeding ponds and remained for longer periods of time than females. In 1978 of 233 captures 195 were males and 38 were females. One hundred thirty-nine of the 147 recaptures that year were males. One male was recaptured 17 days after marking while the longest stay for a marked female was only 5 days (Table 1). In 1980 at Ambystoma Pond 7 of 11 recaptures were male and 4 female. Only two females were recaptured and they had been in the pond 7 and 12 days, respectively. A total of 1122 individuals was captured at this breeding area including 200 females, 2 undetermined and 920 males.

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Notes on the Food Habits of the Long-tail Salamander, Eurycea longicauda, and the Cave Salamander, Eurycea lucifuga

Wynn W. Cudmore
Indiana State University, Terre Haute, Indiana 47809
and
David Rubin
Department of Biology
Central State University, Wilberforce, Ohio 45384

Introduction

It is generally believed that adult salamanders take any prey of appropriate size (Hairston 1949) and that their diets are determined largely by availability of food items (Hamilton 1932). There are few quantitative studies available on the food habits of the long-tail salamander, *Eurycea longicauda*, and the cave salamander, *E. lucifuga*. Minton (1972) reported small terrestrial insects and spiders as the major foods of *E. longicauda* in Indiana, with other arachnids, centipedes, snails and annelids eaten occasionally. Anderson and Martino (1967) described the food habits of *E. longicauda* as being "determined both by availability and preference with some foods taken in proportion to their abundance while others were either not eaten or taken at far below their relative abundance." Coleoptera, spiders, Hymenoptera and isopods were the major foods of individuals from pond margins. Hutchinson (1958) reported heleomyzid flies to be the major foods for small samples of both *E. longicauda* and *E. lucifuga* from in or near caves. Neither species showed predilection to a particular type of food. Peck (1974) provided detailed data on the foods of *E. lucifuga* and found this species to feed on a broader range of food items than *Plethodon glutinosus* in caves. Mycetophilid and heleomyzid flies were the major foods.

The purpose of the present study is to provide additional quantitative information on the food habits of these two species and to investigate seasonal and size relationships.

Methods

Indiana specimens were either collected in Harrison and Crawford counties or taken from the Indiana State University herpetology collection. Museum specimens represented collections primarily from west-central and southern Indiana. All Ohio specimens were collected 2 miles NE of Xenia, Greene County. Salamanders were preserved in 10% formalin and later transferred to alcohol. Snout-vent length was recorded for each individual. Stomachs were removed, examined in water, and their contents were identified using a dissecting microscope. Percent volume for each food was estimated. Data were then summarized as percent volume and percent frequency for Indiana and Ohio separately.

Results and Discussion

Isopods (24.3% volume), spiders (14.3% volume) and adult Diptera (9.8% volume) were the major foods of Indiana longtail salamanders (Table 1). Although Collembola occurred in 34.4% of those stomachs containing food, their contribution to total volume was relatively small at 4.0%. Isopods were terrestrial in nature, spiders were mainly small to medium-sized lycosids and most dipterans were members of the Acalyptratae. Gastropods were represented by small snails and

TABLE 1. *Stomach contents of Eurycea longicauda from Indiana. 80 examined, 61 with food; SV range = 23-61 mm, mean = 46.1, SD = 8.36.*

Food	% Volume	% Frequency
Isopoda	24.3	39.3
Araneae	14.3	36.1
Diptera (adult)	9.8	21.3
Gastropoda	6.8	18.0
Coleoptera (adult)	6.6	18.0
Lepidoptera (larvae)	5.8	9.8
Collembola	4.0	34.4
Coleoptera (larvae)	3.7	13.1
Homoptera (adult)	3.2	9.8
Hemiptera (adult)	2.4	11.5
Diptera (larvae)	2.0	4.9
Orthoptera	2.0	4.9
Hymenoptera	1.7	11.5
Ephemeroptera	1.6	1.6
Diplopoda	1.6	3.3
Earthworm	1.5	1.6
Chilopoda	0.7	4.9
Shed skin	0.7	1.6
Lepidoptera (adult)	0.4	3.3
Pseudoscorpion	0.1	1.6
Unidentified insect	4.6	11.5
Vegetation	1.2	11.5
Detritus	1.1	13.1
	100.1	

slugs (Limacidae). Ohio longtail salamanders (Table 2) fed primarily on Collembola (20.0% volume), adult Diptera (14.5% volume), isopods (11.3% volume) and

TABLE 2. *Stomach contents of Eurycea longicauda from Greene County, Ohio. 81 examined, 72 with food; SV range = 23-57 mm, mean = 41.3, SD = 9.31.*

Food	% Volume	% Frequency
Collembola	20.0	54.2
Diptera (adult)	14.5	27.8
Isopoda	11.3	22.2
Diptera (larvae)	10.8	19.4
Araneae	7.8	18.1
Gastropoda	4.0	6.9
Earthworm	3.6	6.9
Coleoptera (adult)	3.4	6.9
Shed skin	3.1	4.2
Coleoptera (larvae)	2.6	8.3
Orthoptera	2.5	2.8
Homoptera	2.0	9.7
Chilopoda	1.4	1.4
Acarina	1.2	5.6
Plecoptera	1.0	1.4
Hymenoptera	0.9	5.6
Trichoptera (larvae)	0.9	1.4
Hemiptera	0.6	4.2
Unidentified	4.2	11.1
Vegetation	1.9	6.9
Unidentified insect larva	1.4	1.4
Unidentified insect	1.1	4.2
	100.2	

TABLE 3. *Stomach contents (in % volume) of Eurycea longicauda from Indiana categorized by season.*

Food	April-May (N = 21)	June-Aug (N = 25)	Sept-Nov (N = 15)
Araneae	16.5	15.3	9.5
Isopoda	15.0	17.4	48.7
Diptera	21.1	13.1	1.3
Coleoptera	10.5	7.2	— —
Gastropoda	9.5	8.6	0.2
Homoptera	6.5	2.2	— —
Collembola	5.8	3.2	2.6
Coleoptera (larvae)	5.2	1.8	4.7
Diptera (larvae)	5.0	0.8	— —
Lepidoptera (larvae)	2.4	7.0	8.7
Diplopoda	— —	— —	6.3
Orthoptera	— —	5.0	— —

dipterous larvae (10.8% volume). Collembola occurred in over half of those stomachs containing food and unlike Indiana specimens, contributed a major portion of the total volume. Dipterans were mostly very small and included members of the families Phoridae, Mycetophilidae, Tipulidae and Culicidae. Coleoptera from both Indiana and Ohio were primarily staphylinids.

For *Eurycea longicauda*, items accounting for 5% or more of total volume in at least one season are categorized by season for Indiana (Table 3) and for Ohio (Table 4). In both collections adult Diptera were important in spring and summer but decreased dramatically in fall. Collembolans were approximately twice as important in spring as in summer or fall. Perhaps they become less important as a wider variety of foods becomes available. Spiders are the only item that maintained a relatively high importance throughout the year for both collections. Dipterous larvae were approximately twice as important in fall as in other seasons for Ohio individuals. Although Indiana *E. longicauda* showed strong seasonal utilization of isopods (primarily fall) and gastropods (spring-summer), the same trends were not found in Ohio.

Feeding habits of larger longtail salamanders tended to be more diverse than those of smaller individuals. For large Ohio *E. longicauda* (SV 43-57 mm), there were 17 categories of identifiable items and the top 6 accounted for 66.0% of

TABLE 4. *Stomach contents (in % volume) of Eurycea longicauda from Green County, Ohio, categorized by season.*

Food	April (N = 33)	June (N = 16)	Sept-Oct (N = 23)
Collembola	28.6	11.0	13.9
Diptera	18.6	19.5	5.0
Isopoda	14.5	3.1	12.2
Diptera (larvae)	8.5	7.5	16.5
Shed skin	6.7	— —	— —
Earthworm	6.4	3.1	— —
Araneae	6.2	10.0	8.5
Coleoptera	0.9	5.9	5.2
Coleoptera (larvae)	— —	8.5	2.2
Orthoptera	— —	6.3	3.5
Gastropoda	— —	— —	12.4
Homoptera	— —	— —	6.3

total volume, while there were 13 categories of identifiable items taken from small Ohio *E. longicauda* (SV 23-37 mm) with the top 6 accounting for 79.1% of total volume. Although few Indiana individuals were small, a single food category made up 100% of the total volume in 6 of 8 containing food. Only 4 of 43 (9.3%) of large salamanders contained a single category of food. Shed skin made up only a small portion of total volume but was found only in larger salamanders (SV = 47,48,52, and 55 mm). Whitaker and Rubin (1971) found shed skin to be more frequent in larger size classes of *Plethodon jordani*. They proposed that although smaller salamanders would gain more by consuming shed skin since it would be a larger proportion of their total mass, it may be too large for them to consume.

Sixteen *E. longicauda* larvae, all taken in June, were examined. Eleven contained food. Chironomid larvae were the major food at 87.3% volume while coleopterous larvae and Collembola were less important. Caldwell and Houtcooper (1973) found chironomid larvae to be the major food item of *Eurycea bislineata* larvae in March and August from Vigo County, Indiana.

Cave salamanders were collected primarily from localities in Harrison and Crawford counties in Indiana. Most were collected from the twilight zone of caves; however, some were from open limestone areas. Forty cave salamanders were examined, 15 of which contained no food. The data for the remaining 25 are given in Table 5. Adult dipterans (28.7% volume), spiders (18.5% volume), and Coleoptera (18.1% volume) were the major food items. Dipterans were represented by acalypterate families, and spiders were mostly lycosids. Cave salamanders showed no seasonal shift in food habits as all major foods were well represented in each season. While isopods and springtails were the most important foods for Indiana and Ohio *E. longicauda* respectively, these foods were relatively unimportant in this sample of *E. lucifuga*. Adult Diptera and spiders were the only items that contributed significantly to the foods of both species. In a study of the foods of *Eurycea bislineata*, Hamilton (1932) found Coleoptera, spiders, isopods,

TABLE 5. *Stomach contents of Eurycea lucifuga from Indiana. 40 examined, 25 with food; SV range = 30-71 mm, mean = 56.8, SD = 8.26*

Food	% Volume	% Frequency
Diptera (adult)	28.7	48.0
Araneae	18.5	36.0
Coleoptera (adult)	18.1	28.0
Earthworm	8.0	8.0
Lepidoptera (larvae)	7.8	16.0
Shed skin	3.0	8.0
Hymenoptera	2.6	12.0
Isopoda	2.4	8.0
Orthoptera	2.0	8.0
Chilopoda	2.0	4.0
Diptera (larvae)	1.4	4.0
Collembola	1.0	12.0
Hemiptera (adult)	0.4	4.0
Coleoptera (larvae)	0.4	4.0
Acarina	0.3	8.0
Pseudoscorpion	0.2	4.0
Lepidoptera (adult)	0.2	4.0
Psocoptera	0.1	4.0
Unidentified insect	1.5	16.0
Vegetation	1.4	12.0
	100.0	

mayflies and Diptera to be the major foods. Of these, spiders, isopods and Diptera were important foods of *E. longicauda.*

Indications from the literature and the present study are that *E. lucifuga* feeds primarily on adult Diptera while *Eurycea longicauda* tends to be more versatile in food habits with major foods being quite different in several studies. This likely reflects broader habitat tolerances of *E. longicauda,* but an actual preference for foods on the part of *E. lucifuga* cannot be entirely ruled out at this time.

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Present Distribution of Beaver in Indiana

Gary Langell
Indiana Department of Natural Resources
Division of Fish and Wildlife
Bloomington, Indiana 47401

Introduction

In 1955, 20 years after beaver reintroduction efforts began, Brooks (2) attempted to delineate the distribution of beaver in Indiana. The principal beaver range was determined to be primarily associated with the Kankakee River drainage basin in northwestern Indiana (Figure 1). Since then, beaver range has expanded. The purpose of this report is to provide current data on the distribution of beaver in Indiana.

Methods

In August of 1981, questionnaires were sent to District Wildlife Biologists of the Indiana Department of Natural Resources (IDNR) requesting the locations of all known beaver colonies within their district and the general habitat associated with each colony. Biologists contacted IDNR Conservation Officers within their respective districts and then added other known sightings. In many instances, officers contacted local trappers to obtain more complete information. Beaver sightings, beaver dams and lodges, beaver damage complaints, and beaver cuttings were used to denote colony locations. Miles of streams per county were measured from a scaled map (4) using a Keuffel and Esser map measure instrument.

Results

A total of 1,218 beaver colonies was reported in 76 of 92 possible counties. No colonies were reported from a 15 county area in east central and southeastern Indiana (Figure 2). Over one-half of the reported colonies occurred in southern Indiana, with the largest concentration in the southwest (Table 1). Almost two-thirds of the colonies were located on rivers and streams, with the majority being associated with the Ohio, Wabash and Kankakee Rivers and their major tributaries. Water impoundments resulting from surface coal mining in southwestern Indiana were the second most commonly used habitat type, followed by ditches, ponds, and lakes. Less than one percent of the total colonies were associated with reservoirs. Over three-fourths of the colonies associated with lakes occurred in the natural lakes region of northern Indiana. Numbers of beaver per county were positively correlated with miles of streams per county ($r = 0.41$, $P > 0.05$).

Discussion

Beaver have substantially expanded their range in Indiana since 1955. Their distribution is primarily associated with the major river systems and their tributaries and encompasses all but 15 counties in east central and southeastern Indiana. Although isolated beaver colonies may exist in east central and southeastern Indiana, their presence is not known at this time.

Reasons for the absence of beaver in the east central and southeastern portions of the state are not known. Most of Indiana's major streams originate in east central Indiana. Water levels are variable and stream flow is rapid (3). The

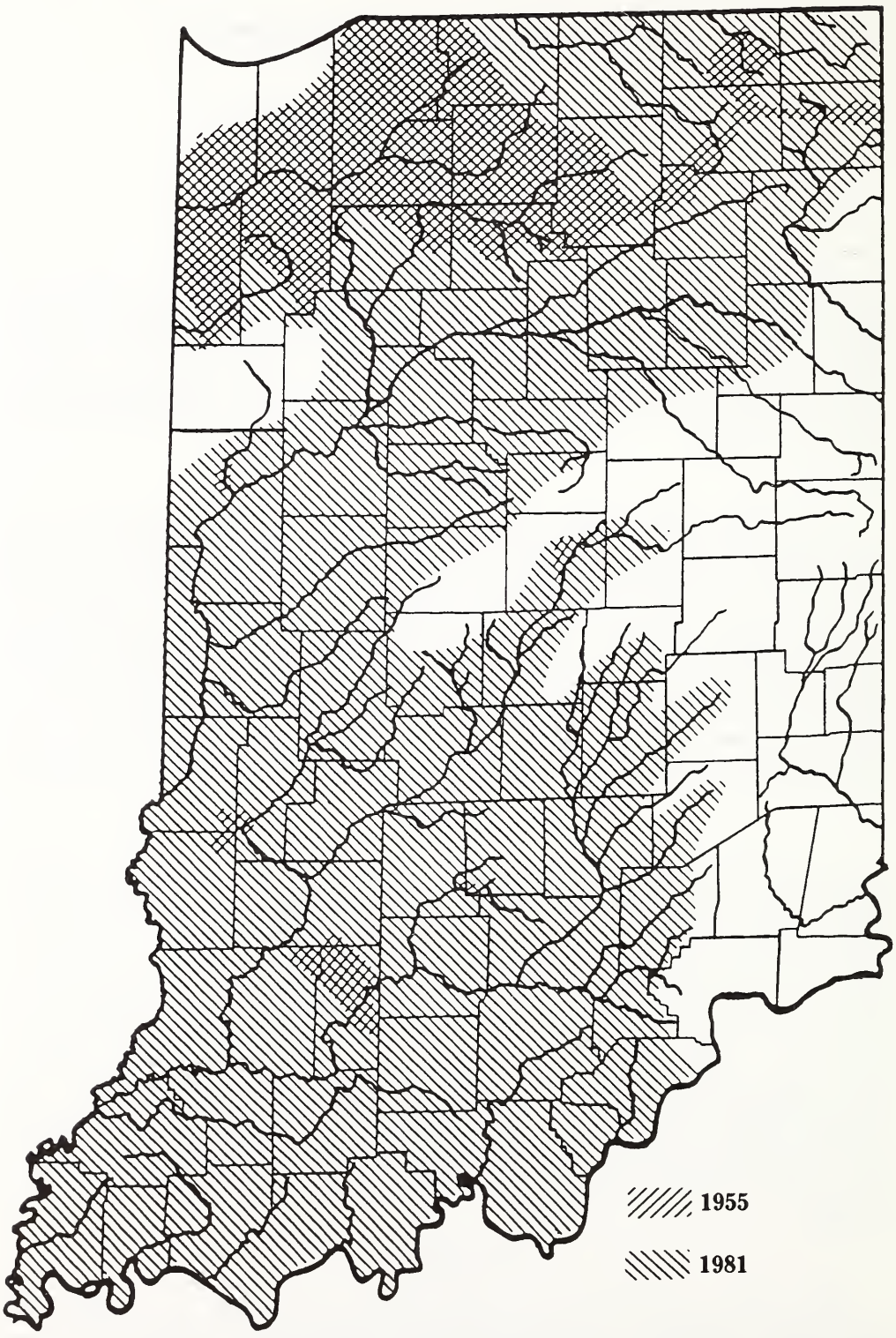

FIGURE 1. *Distribution of beaver in Indiana in 1955 and 1981.*

combination of these two variables may deter colonization. Flood control reservoirs on the Mississenewa and Salomonie Rivers, the major drainage systems for the east central region, may also impede upstream colonization. Colonization of these rivers is limited to those portions downstream from the water control

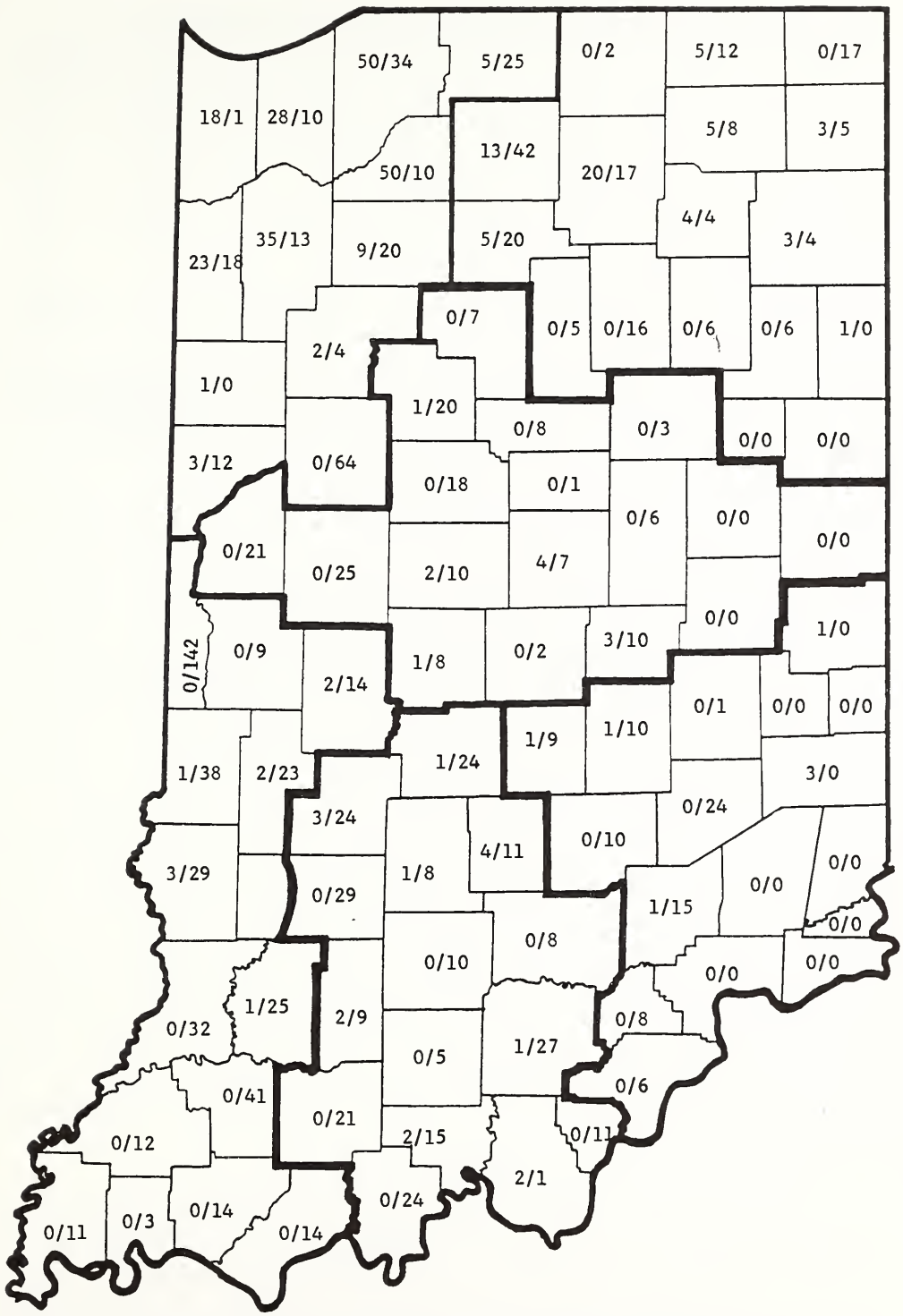

FIGURE 2. *Distribution of beaver colonies by counties in Indiana (1955/1981).*

structures. Transplanting beavers above the reservoirs might aid in range expansion.

In southeastern Indiana, terrain may be inhibiting colonization. This region is separated from the other drainage basins of southern Indiana by the Dearborn Uplands, a series of narrow winding ridges with steep slopes (1). The uplands

TABLE 1. *Number of beaver colonies reported by zone and habitat type.*

	NW	NE	C	SW	SC	SE	TOTAL	PERCENT
Streams	144	84	111	255	150	59	803	65.0
Strip Pits	0	0	0	110	5	0	115	9.4
Ditches	39	10	25	31	2	1	108	8.9
Ponds	15	20	6	25	34	0	100	8.2
Lakes	13	49	2	1	12	3	80	6.6
Reservoirs	0	1	2	0	9	0	12	1.0
TOTALS	211	164	146	422	212	63	1,218	— — —
PERCENT	17.3	13.5	12.0	34.6	17.4	5.2	— — —	100.0

create an isolated drainage basin that flows eastward into Ohio. Streams of this area characteristically have variable water levels and rapid streamflow (3).

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Recent Records and Status of Amphibians and Reptiles in Indiana

SHERMAN A. MINTON, Indiana University School of Medicine, Indianapolis, Indiana 46223
JAMES C. LIST, Ball State University, Muncie, Indiana 47306
and
MICHAEL J. LODATO, 925 Park Plaza Drive, Evansville, Indiana

Introduction

The purpose of this paper is to review the status of Indiana amphibians and reptiles since the publication of the last comprehensive work on the group (4). Our principal goal is to present observations on changes in abundance of species over the past decade and to report new records representing extensions of range or occurrence at new sites within the presumed Indiana range. We have accumulated a large number of such records, most of them supported by preserved specimens or color photographs. Most of the identifications in this account have been made by one of us; however in some cases we have accepted reports of reliable and well qualified observers. While most of the evidence we present is anecdotal, we are long-term Indiana residents who have spent a significant part of our professional lives making field observations of the state herpetofauna and advising and directing others in faunal studies. We feel that impressions gained over the years may partially compensate for lack of quantitative data on populations we have observed. A secondary goal is to recognize recent changes in taxonomy and nomenclature that have affected Indiana species of amphibians and reptiles.

Finally, by inference as well as statement, this review offers a comment on the environment that provides the habitat for the Indiana herpetofauna. The past two decades have witnessed a growing concern with environmental change and its effect on local biotas. In many ways, amphibians and reptiles are sensitive indicators of such environmental change. Many have exacting requirements for moisture, temperature, shelter, and food during all or part of their life cycles. Nearly all have poor powers of dispersal, spending their entire lives within an area of a few hectares, sometimes much less. Many are quite sensitive to chemical toxicants such as pesticides, herbicides, and industrial wastes. While much remains to be learned, the ecology of many common North American species is sufficiently well known to allow them to serve as indicators of environmental deterioration.

Accounts of Species

Cryptobranchus a. alleganiensis (Hellbender): A study of Indiana populations of this species by William Kern has confirmed its presence only in Blue River where it has been found as far as southern Washington County. A specimen was collected about 1963 in the Whitewater River, Franklin County.

Necturus m. maculosus (Mudpuppy): Small populations apparently persist in most major streams and natural lakes in Indiana, but there is little new information. New county records: Brown, Delaware, Lagrange, Randolph, Whitley.

Ambystoma jeffersonianum (Jefferson's Salamander): New populations have been in Jennings County (Muscatatuck Wildlife Refuge) and Perry County (1.5 miles ne. Oriole). Females from Bartholomew County (4 miles sw. Columbus; Taylorsville) may belong to this species or the triploid *platineum*.

Ambystoma laterale (Blue-spotted Salamander): No new Indiana populations of this species have been identified.

Ambystoma platineum, A. tremblayi (Triploid all-female populations of the *jeffersonianum* complex): Populations identified on the basis of erythrocyte size occur in Wayne Township, Allen County; Bryan Nature Preserve, Clinton County; and Moraine Nature Preserve, Porter County. A St. Joseph County population (Kern Road, 6 miles sw. South Bend) is tentatively assigned to this group since a sample of 9 individuals contains no males. A single immature specimen from 2 miles south of Mentone, Kosciusko County is assigned to this group on the basis of erythrocyte size. The Boone County colony of *platineum* (4 miles e. of Whitestown) has been destroyed by habitat modification. The Clinton County specimens are typical of *platineum* and the St. Joseph County specimens typical *tremblayi*, however the Allen and Porter County specimens cannot definitely be assigned to either form but seem closer to *platineum*.

Ambystoma texanum (Small-mouth Salamander): This species is regularly associated with *jeffersonianum* complex triploids in breeding ponds as well as with typical diploid *jeffersonianum*. New county records: Franklin, Henry, Jennings, Montogomery, Randolph, Warrick.

Ambystoma maculatum (Spotted Salamander): No significant new information. New county records: Franklin, Henry, Jennings, Laporte, Vanderburgh.

Ambystoma t. tigrinum (Eastern Tiger Salamander): Apparently more plentiful in extreme southern Indiana than previously believed. New county records: Clark, Clinton, Floyd, Harrison, Henry, Lagrange, Randolph, Warrick.

Ambystoma opacum (Marbled Salamander): No significant new information or county records. Northern and eastern limits of the state range are poorly known.

Notophthalmus viridescens (Newt): New county records: Crawford, Delaware, Franklin, Henry, Perry, Porter, St. Joseph, Switzerland. The Crawford and Perry County specimens, mostly efts, are typical of the eastern subspecies, *N.v. viridescens*; Delaware, Henry, and St. Joseph County specimens are more typical of the central subspecies, *N.v. louisianensis*.

Desmognathus f. fuscus (Northern Dusky Salamander): Locally abundant. Recent new records extend the range into southern Henry County and eastern Lawrence County.

Hemidactylium scutatum (Four-toed Salamander): A colony of this uncommon salamander is located in a woodland swamp 1.5 miles south of Springport, Henry County and an attempt is being made to protect this site. Other new Indiana localities are Pigeon River Fish and Wildlife Area, Lagrange County and an unspecified locality in northwestern St. Joseph County.

Plethodon g. glutinosus (Slimy Salamander): A generally common woodland salamander in southern Indiana. No new county records.

Plethodon cinereus (Redback Salamander): A common small vertebrate in most Indiana woodland. At the southern end of the Knobstone Escarpment in Floyd and Clark Counties, this species seems to have increased in numbers since about 1975, while the closely related *P. dorsalis* has decreased. During the 1946-55 period when one of us (S.A.M.) was collecting regularly near New Albany, *P. dorsalis* was repeatedly listed as numerous with as many as 5 being found under one rock, while only 2 individuals of *P. cinereus* were mentioned in a nine-year period. One of these was collected 15 May 1948 near Renn's Spring on Bald Knob along with 8 *P. dorsalis*. At this same locality on 27 April 1979, 10 *cinereus* and 2 *dorsalis* were found and on 21 February 1981, about 12 *cinereus* and 1 *dorsalis*. New

county records: Clark, Delaware, Dubois, Grant, Perry, Posey, Washington, Warrick.

Plethodon dorsalis (Zigzag Salamander): No significant new information except as noted under *P. cinereus*. New county records: Greene, Posey.

Plethodon richmondi (Ravine Salamander): A recent record from Fayette County narrows the gap between Ohio valley populations and those in Wayne and Henry Counties.

Pseudotriton r. ruber (Eastern Red Salamander): First reported from Indiana by Wilson (9) on the basis of two specimens collected in Floyd County 4 miles northwest of New Albany. No others have been reported. The collection area is now largely suburban, but the species may still survive in small, spring-fed streams.

Eurycea b. bislineata (Two-lined Salamander): Common in many rocky brooks of southern Indiana. New county records: Delaware, Grant, Huntington, Ohio.

Eurycea l. longicauda (Long-tailed Salamander): A record from bluffs near West Franklin on the Posey-Vanderburgh County line extends the range into the Wabash lowland. Other new county records: Dubois, Franklin, Greene, Jackson.

Eurycea lucifuga (Orange Cave Salamander): Associated with *E. longicauda* at the West Franklin locality.

Siren intermedia nettingi (Western Lesser Siren): Locally and seasonally common in extreme southwestern Indiana; recently recorded at Jasper-Pulaski State Fish and Wildlife Area. New county records: Jasper, Posey, Vanderburgh, Warrick.

Scaphiopus h. holbrooki (Eastern Spadefoot): Indiana distribution very spotty and activity unpredictable. The only new record is a single adult female collected near Oriole, Perry County.

Bufo americanus (American Toad): Presence of two forms of the American toad in Indiana has been recognized for several decades. One is reddish-brown in life with little dorsal spotting, light ventral pigmentation, and small size (males 50-65 mm body length, females 60-75). The other is yellowish to greyish brown with prominent dark spots, heavy ventral dark pigmentation, and large size (males 65-80 mm body length, females 75-87). Differences are most apparent when specimens from the northwestern muck prairie are compared with individuals from the southern central hill country. The former belong to the large, spotted form, the latter to the small reddish form. Toads from the central and northeastern part of the state are generally intermediate in size, pattern, and ventral pigmentation, however those from the Indiana Dunes and northwestern sand prairie tend to be small with little dorsal spotting but heavy ventral pigmentation. The small, light-bellied toads of southern Indiana may represent the subspecies *charlesmithi* and the larger toads of northern and central Indiana the nominate subspecies, however ranges and zones of intergradation remain to be defined. There are still no records of *americanus* from the lower Wabash valley counties of Indiana and Illinois. New county records: Clark, Crawford, Decatur, Delaware, Henry, Howard, Jasper, Jay, Madison, Randolph, Shelby, Spencer.

Bufo woodhousei fowleri (Fowler's Toad): The degree of hybridization between Fowler's toad and the American toad in Indiana remains undetermined. Ottys Sanders (pers comm.) notes subtile differences between *fowleri* of Indiana and Illinois, the former being more brownish with less divergent interorbital crests, features that could indicate the genetic influence of *americanus*. New county records: Blackford, Crawford, Delaware, Franklin, Jay.

Acris crepitans blanchardi (Cricket Frog): It is our impression that this species

has greatly decreased in numbers in central Indiana. In southwestern Indiana it remains plentiful about the margins of borrow pits and shallow strip mine lakes. New county records: Franklin, Lagrange, Perry, Randolph, Shelby.

Pseudacris triseriata (Striped Chorus Frog): This small frog, extremely plentiful throughout most of the state in the 1940's and 1950's, has decreased greatly in numbers in the last decade. In 1966 there were 10 populations of *Pseudacris* in marshy spots within a 3-mile radius in northesastern Marion County; in 1981 there were none. In some cases, habitats have been destroyed, but in others at least marginal habitat remains. On the night of April 2, 1982, one of us (S.A.M.) drove 69 miles on secondary roads in Clay, Owen, and Putnam Counties and heard a single *Pseudacris* at close range, perhaps three others at a distance. Many *Hyla crucifer* and *Bufo americanus* were calling. New county records: Delaware, Henry, Lagrange, Randolph, Steuben.

Hyla c. crucifer (Spring Peeper): Seems to be increasing in numbers, particularly where *Pseudacris* has become uncommon. It is thriving near old strip mine spoil areas that were reclaimed by reforestation. New county records: Delaware, Henry, Jennings, Randolph, St. Joseph, Warrick.

Hyla versicolor and *H. chrysoscelis* (Eastern Grey Treefrog): Since these species can be distinguished only by voice and chromosome count, their Indiana ranges are poorly known. *H. chrysoscelis* seems to be the dominant form in the southern part of the state, ranging north at least to Delaware County; *versicolor* is known only from Indianapolis northward. New county records: Delaware, Franklin, Henry, Lagrange.

Rana clamitans melanota (Green Frog): Generally common in woodland. New country records: Delaware, Franklin, Henry, Jay, Randolph.

Rana catesbeiana (Bull Frog): Common in most permanent streams, ponds and lakes. New county records: Delaware, Franklin, Henry, Jackson, Jay, Jennings.

Rana areolata circulosa (Crawfish Frog): Some colonies in Vanderburgh and Warrick Counties have been destroyed by surface mining, drainage, and urban expansion. A report from near Monrovia, Morgan County extends the range slightly but requires confirmation.

Rana pipiens (Northern Leopard Frog), *R. utricularia* (Southern Leopard Frog), and *R. blairi* (Plains Leopard Frog): Minton (4) treated *pipiens* and *sphenocephala* (*utricularia*) as subspecies. Map 31 shows roughly the distribution of these two species in Indiana save that the zone of intergradation is actually a zone of overlap. The recently described *Rana blairi* is definitely known in Indiana only from Benton and Vigo Counties. Study of leopard frog systematics is particularly needed in southeastern and west central Indiana. Leopard frogs are among the formerly plentiful amphibian species that have decreased markedly in numbers since about 1970. This is particularly true of the northern species.

Rana palustris (Pickerel Frog): No significant new information. New county records: Jennings, St. Joseph.

Rana sylvatica (Wood Frog): One of us (J.C.L.) has noted the disappearance of a population of these frogs in a 100-acre woods in Delaware County. The last specimen was seen in 1959. The reason for their disappearance is unknown.

Chelydra serpentina (Snapping Turtle): Commercial turtle hunters report a marked decrease in snapping turtles in central Indiana since 1970. New county records: Delaware, Henry, Lagrange, Randolph.

Macroclemys temmincki (Alligator Turtle): A newspaper account with photographs

reports capture of two large specimens in a slough off the Ohio River in western Kentucky in May, 1980. This gives reason to believe this huge turtle may still be found in the lower Wabash and adjacent Ohio.

Sternotherus odoratus (Musk Turtle): No significant new information. New county records: Delaware, Henry, Madison, Steuben, Vanderburgh.

Kinosternon s. subrubrum (Eastern Mud Turtle): Road-killed turtles were collected in Posey County near Hovey Lake in 1974 and near Griffith in 1976. The Indianapolis Children's Museum had a specimen allegedly collected in Newton County in 1979.

Clemmys guttata (Spotted Turtle): This small, attractive turtle is rare in Indiana largely as a result of destruction of its marsh and bog habitat and the collection of specimens for the pet trade. It has recently been reported from near Delphi, Carroll County, Pigeon River Fish and Wildlife Area, Lagrange County, and Potato Creek Recreation Area, St. Joseph County.

Emdoidea blandingi (Blanding's Turtle): Status similar to that of the spotted turtle but seems less severely threatened. New county records: St. Joseph, Tippecanoe.

Terrapene c. carolina (Eastern Box Turtle): Distinctly less plentiful today than in the 1950's. New county records: Henry, St. Joseph.

Terrapene o. ornata (Ornate Box Turtle): Few recent records, all from relict sand prairies in northwestern Indiana.

Graptemys geographica (Map Turtle): No significant new information. New county records: Delaware, Elkhart, Henry.

Graptemys pseudogeographica (Midland Sawback Turtle) and *G. ouachitensis* (Ouachita Sawback Turtle): Vogt (7) considers these turtles distinct species. In Indiana they appear to be sympatric, although further studies of distribution and ecology are badly needed. Ewert (2) has reported extension of the Indiana Range of *ouachitensis* in the White River drainage to Columbus and in the Ohio to Jeffersonville. New county records: Bartholomew, Clark, Jackson, Warrick, Vanderburgh.

Chrysemys picta marginata (Midland Painted Turtle): Common in ponds throughout the state. New county records: Henry, Jennings, Randolph, St. Joseph.

Chrysemys scripta elegans (Red-eared Turtle): Recent records (2, 6) indicate this turtle may occur in all the Ohio valley counties of Indiana and throughout most of the White River system as well. New county records: Bartholomew, Floyd, Harrison, Owen, Perry, St. Joseph, Shelby, Vanderburgh.

Chrysemys concinna hieroglyphica x *P. floridana hoyi* (Hieroglyphic Turtle): This interesting turtle population apparently persists in extreme southwestern Indiana. In July, 1972 an adult male was collected at the junction of Black River with the Wabash near New Harmony. In 1973, the shell of an adult was found in Point Township, Posey County.

Trionyx s. spinifer (Eastern Spiny Softshell): No significant new information. New county records: Jay, Randolph, Shelby.

Trionyx m. muticus (Midland Smooth Softshell): Ewert (2) reported extension of the species range in the east fork of the White River. New county records: Bartholomew, Jackson, Shelby.

Sceloporus undulatus hyacinthinus (Northern Fence Lizard): Generally plentiful in open woods and forest edge in southern Indiana.

Ophisaurus a. attenuatus (Western Ophisaur): Always apparently uncommon in Indiana, this lizard persists in the northwestern sand prairies. Recent records include 4 individuals at Jasper-Pulaski State Fish and Game Area in 1971, a road-kill in northern Jasper County in May, 1977, and an individual photographed in Clark and Pine Dune and Swale in Lake County in April, 1981.

Cnemidophorus s. sexlineatus (Six-lined Racerunner): Finding of a small colony near Owensville, Gibson County confirms southwestern Indiana records about a century old. A sight record at a gravel prairie remnant near Wea Creek, Tippecanoe County evidently indicates a small relict population in the till plains. On the knobs north of New Albany, the racerunner is decreasing in numbers apparently due to increased growth of vegetation on the formerly almost bare claystone slopes. At the northernmost colony of the Floyd-Clark County line, no lizards have been identified since 1971. None have been seen at the Dug Knob colony since about 1950. The two southern colonies, Renn's Knob and Moser Knob, still have populations apparently in part because of construction of a large power line that keeps sections of the area relatively open.

Scincella laterale (Ground Skink): New records include three localities in Perry County.

Eumeces fasciatus (Five-lined Skink): Common in the southern region; rare north of Indianapolis. New county records: Allen, Delaware, Dubois, Fayette, Franklin, Jay, Jennings, Henry, Madison.

Eumeces laticeps (Broad-headed Skink): Widely distributed but uncommon. No new county records.

Nerodia sipedon (Banded Watersnake): Generally plentiful throughout Indiana. An unusual specimen was collected near Hovey Lake, Posey County by Robert Herrington in 1973. It was dark yellow with about 25 unbroken sooty black crossbands and reddish ventral spots arranged in parallel rows. *N. sipedon* is not a common snake in Posey County, but the few I have seen have been quite typical of *N. s. pleuralis* of southern Indiana and adjacent Kentucky and Illinois and quite different from the Herrington specimen which resembles a hybrid between *N. sipedon* and *N. fasciata*. New county records: Delaware, Dubois, Henry, Jay, St. Joseph.

Nerodia erythrogaster neglecta (Northern Copperbelly): Destruction of swamp forest has virtually eliminated this species in northeastern Indiana; the most recent report is of a specimen taken "northeast of Angola" in 1974. A fairly large population remains on the Muscatatuck Wildlife Refuge in south central Indiana. There are several colonies of the species in extreme southwestern Indiana. New county records: Jennings, Warrick.

Nerodia r. rhombifera (Diamondback Watersnake): No significant new information. New county records: Pike.

Regina septemvittata (Queen Snake): No significant new information. New county records: Delaware, Jay, Henry, Randolph.

Clonophis kirtlandii (Kirtland's Snake): This little snake has disappeared from most urban and suburban habitats in the Indianapolis area where it was once common. Fewer than a dozen have been found in the last decade. A record from Valeene, Orange County extends the range about 55 miles south on the Mitchell Plain. Additional new county record: Blackford.

Storeria dekayi wrightorum (Midland Brown Snake): No significant new information. New county records: Allen, Jennings, Lagrange, Laporte, Madison, Perry, Washington.

Storeria o. occipitomaculata (Northern Redbellied Snake): Records from Griffith Prairie, Lake County (5), Grabill, Allen County, and Kingsbury State Fish and Wildlife Area, Laporte County indicate a wide albeit spotty distribution in northern Indiana. Northern Indiana specimens do not differ significantly in scale counts and pattern from those of southern Indiana. New southern county records: Scott, Spencer.

Virginia valeriae elegans (Western Earth Snake): No significant new information. New county records: Clark, Monroe, Perry.

Thamnophis s. sirtalis (Eastern Garter Snake): No significant new information. New county records: Dearborn, Madison, Perry.

Thamnophis r. radix (Plains Garter Snake): No significant new information.

Thamnophis butleri (Butler's Garter Snake): Recorded from Kendallville, Noble County on the basis of specimens supplied by a commercial dealer in 1973. The urban colony in Kokomo probably has been destroyed. Only *Storeria dekayi* was found there in the spring of 1976, and no snakes were found in the spring of 1979.

Thamnophis sauritus (Ribbon Snake): Specimens from Coffee Bayou, Gibson County and the Muscatatuck Wildlife Refuge have been identified as the Eastern Ribbon Snake (*T. s. sauritus*) while specimens from the fish hatcheries near Martinsville, Morgan County, Warren Township, Putnam County, and the Pigeon River Fish and Wildlife Area are typical of the Northern Ribbon Snake (*T. s. septentrionalis*).

Thamnophis p. proximus (Western Ribbon Snake): There have been no definite Indiana records of this species since 1963. Its status requires reevaluation.

Coluber constrictor (Racer): Most recent workers recognize the blue racer of northern Indiana and adjacent Illinois, Ohio, and Michigan as a distinct subspecies, *C. c. foxi*. The area of intergradation between the blue and black racer in Indiana is slightly wider than shown in Minton (4, map 67). New county records: Delaware, Henry, Jay.

Elaphe obsoleta (Rat Snake): Acquisition of additional specimens from extreme southwestern Indiana causes us to refer snakes of Posey, Vanderburgh, Gibson, and Warrick Counties to the subspecies *spiloides* as currently defined. Intergradation with *E. o. obsoleta* occurs throughout most of southern Indiana north through Parke, Morgan, and Dearborn Counties. New county records: Delaware, Henry, Pike, St. Joseph, Shelby, Warrick.

Elaphe v. vulpina (Western Fox Snake): Records from Clinton County and north central St. Joseph County slightly extend the eastern limits of the state range.

Pituophis melanoleucus sayi (Bull Snake): No significant new information.

Lampropeltis c. calligaster (Prairie Kingsnake): The virtual absence of records from the Ohio valley counties of southwestern Indiana is difficult to explain. Anthony Wilson reported collecting a specimen near New Albany about 1972.

Lampropeltis getulus niger (Black Kingsnake): Records from Lovett Township, Jennings County represent a slight extension of the range to the northeast.

Lampropeltis triangulum (Milk Snake): Williams (8) considers most of southern Indiana a zone of intergradation between the northern milk snake (*L. t. triangulum*) and the red milk snake (*L. t. syspila*). Recent collections from southern Indiana indicate the population in Harrison and eastern Perry Counties is intergrade rather than *syspila*. However, typical specimens of *syspila* have been found in Knox, Martin, and Orange Counties. The milk snake is rare in extreme southwestern Indiana. New county records: Delaware, Henry, Jay, Jennings, Lagrange, St. Joseph, Whitley.

Cemophora coccinea copei (Scarlet Snake): No additional specimens of this snake have been collected. Areas in Floyd County where specimens were taken are now suburban, however this is a species that adapts well to disturbed habitats.

Opheodrys aestivus (Rough Green Snake): Found regularly in the southern hill country but no longer plentiful. New county record: Daviess.

Opheodrys vernalis blanchardi (Smooth Green Snake): A specimen found 9 miles east of Fowler near the Benton-White County line in 1973 represents the most recent Indiana record known to us. There are occasional reports of small green snakes from near Indianapolis, but we have seen no specimens. Holman and Richards (3) reported vertebrae of this species from a cave in southwest Monroe County associated with late Pleistocene mammal remains. Within historic times, *O. vernalis* has been reported from Brown, Knox and Posey Counties, however these records are nearly a century old.

Diadophis punctatus edwardsi (Northern Ringneck Snake): A common small snake in southern Indiana woodland. New county records: Franklin, Jackson, Perry, Wayne.

Tantilla coronata (Crowned Snake): We have not recorded this species from Indiana since 1967, however its habitat in Floyd and Clark Counties does not seem to be seriously disturbed.

Carphophis amoenus helenae (Worm Snake): No significant new information. New county records: Dubois, Franklin.

Heterodon platyrhinos (Eastern Hognose Snake): No significant new information. New county records: Delaware, Fayette, Lagrange, St. Joseph.

Agkistrodon contortrix mokeson (Northern Copperhead): Although numbers seem to be reduced, the Indiana range of the copperhead remains essentially the same. A recent report from Franklin County confirms its presence there. New county records: Gibson, Jennings, Warrick.

Sistrurus c. catenatus (Eastern Massasauga): A report from Carroll County in 1981 indicates persistence of at least one till plains colony. Other reports in the last five years are for Kosciusko, Lagrange, Laporte, St. Joseph and Steuben Counties.

Crotalus h. horridus (Timber Rattlesnake): Rattlesnakes are reported regularly from Brown, Monroe, and Morgan Counties, rarely from other southern Indiana localities. New county records: Harrison, Perry.

Summary

In 1972, 82 species of amphibians and reptiles were reported to inhabit Indiana. Two species have been exterminated apparently within historic time, the mud snake (*Farancia abacura*) about 1900 and the alligator snapping turtle (*Macroclemys temmincki*) about 1935. Both had very small ranges in the southwest tip of the state. Since 1972, five species have been added, one (*Pseudotriton ruber*) by discovery; the others by definition of previously unrecognized sibling species. Five species (*Pseudotriton ruber*, *Rana blairi*, *Chrysemys concinna*, *Cemophora coccinea*, and *Tantilla coronata*) have minute Indiana ranges consisting of small parts of 3 or fewer counties. All are species at the extreme periphery of more extensive continuous ranges. All are still believed to occur in Indiana although *Cemophora* has not been recorded since 1957. Ten species have conspicuously disjunct and probably relictual Indiana ranges; six of these (*Hemidactylium scutatum*, *Kinosternon subrubrum*, *Nerodia erythrogaster*, *Thamnophis butleri*, *Thamnophis proximus*, and *Opheodrys vernalis*) have had their numbers markedly reduced through habitat

modification. Of 7 additional species whose ranges extend but a short distance into Indiana, *Cryptobranchus alleganiensis* has been virtually eliminated within the past 50 years while *Ophisaurus attenuatus* is rare but apparently was never plentiful. Species formerly widespread in Indiana whose numbers have declined markedly in the state since 1950 include *Pseudacris triseriata*, *Rana pipiens*, *Rana areolata*, *Clemmys guttata*, *Emydoidea blandingi*, *Clonophis kirtlandi*, and *Sistrurus catenatus*. The decrease has been marked enough to justify threatened status for *Clemmys* and *Sistrurus*. Less marked but probably significant decreases in numbers have been noted for *Necturus maculosus*, *Chelydra serpentina*, *Terrapene carolina*, *Opheodrys aestivus*, and *Thamnophis sauritus*.

Christiansen (1) estimates that ranges of 69 percent of Iowa's reptiles and amphibians are decreasing and 15 percent are in imminent danger of extinction in the state. Destruction of prairie marshes and woodland ponds and loss of the prairie-forest ecotone have been major factors contributing to the decline of populations. Salamanders, snakes, and lizards are affected more severely than frogs and turtles. Of 57 amphibian and reptile species common to the two states, 33 appear to be faring at least marginally better in Indiana while 4 are better in Iowa. *Opheodrys vernalis* is considered a threatened species in both states; *Nerodia erythrogaster* and *Sistrurus catenatus* are listed as threatened in Indiana, endangered in Iowa.

Note added in proof: In May, 1983 *Agkistrodon piscivorus leucostoma* (Western Cottonmouth) was recorded from Indiana for the first time at Buffalo Bottoms near Jasper, Dubois County. This increases the number of species in the state to 88.

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Role of Tannic Acid on Digestion by Red Squirrels

Eric D. Mould and Kara Eberly
Department of Biology
Saint Mary's College, Notre Dame, Indiana 46556
and
Douglas Duff
Department of Biology
Indiana University, South Bend, Indiana 46634

Introduction

The effects of dietary phenolics (or tannins) have been of considerable interest to nutritionists and ecologists because they apparently reduce nutrient availability and subsequently affect metabolism. Phenolics form both reversible and irreversible chemical complexes with proteins and, to a lesser extent, soluble carbohydrates (46). Formation of these complexes in the gut is potentially deleterious to digestion and/or assimilation regardless of whether the phenolics complex with macromolecules of dietary (31) or endogenous origin (26). Additionally, phenolic complex formation may greatly alter gastric and intestinal secretions (9,12,22,25), and nutrient assimilation may be hindered by the action of phenolics on intestinal epithelium (27). Some phenolics may themselves be hydrolized by digestive enzymes or gut microflora and absorbed (24). Circulating phenolics have been implicated with a wide variety of metabolic dysfunctions including interruption of general protein and lipid metabolism (39), changes in cholestrol metabolism (49), hepatic malfunctions (36), interaction with thiamine (40), alterations in the metabolism of methionine and choline (3), and impairment of reproductive performance (7,38).

Phenolic content has been correlated with reduced palatability to a variety of herbivorous species (11,13,30,34,35), probably as a result of their astringent characteristics when in contact with oral epithelium or subsequent metabolic effects. However, interpretation of herbivore-plant interaction may be significantly complicated by the observation that in some cases phenolics promoted efficient digestion by favorably altering gut pH when acidosis existed. The potential for ruminal or cecal acidosis is increased when grains represent a large portion of the diet (48). In fact, some ungulates may even select bark containing high concentrations of phenolics to control rumen fermentation (Prins—personal communication 1977).

Knowledge of animal adaptations to plant defenses to herbivory are generally lacking. Although some research has been concerned with the response of ruminants to plant secondary compounds (8,15,16,22,25,29), almost nothing is known about the response of wild monogastric herbivores to the ingestion of phenolics. Most nutritional studies of squirrels and other small herbivorous mammals have concentrated on food habits and energy contributions of foods. Certainly the viability of squirrel populations is dependent upon nutritional parameters since their breeding rate depends on the extent of fall mast crop (2,5,19,32,33,37,45). Presumably the biological quality of the crop is an important determinant as well. Forage selectivity and physiological considerations of squirrels apparently involve dynamic interelationships of mineral assimilation (14,43) and energy metabolism (17,21,28,42). Ofcarcik et al. (35) inferred that there exists an inverse relationship of phenolic content and feeding preferences of squirrels. However, no study has

been conducted on any herbivore for the purpose of quantitatively elucidating the digestive effects of phenolics. The multitide of changes in pH and substrate availability in the gut complicate the precise definition of phenolic influence since no quantitative methodologies assure accurate determination of phenolic content of both feed and feces. Such problematic considerations can, however, be avoided if purified diets are utilized to which phenolic content is added in known quantities. This study seeks to determine the quantitative effects of a common phenolic (tannic acid) on the digestive response of captive red squirrels (*Tamiasciurus hudsonicus*) consuming different phenolic concentrations in otherwise equivalent artificial diets.

Methods

Mature male squirrels were live trapped in St. Joseph Co., Indiana and maintained in individual wire cages (45x45x35 cm). Squirrels were fed a balanced rodent ration from Bio-Serv. Inc. (18.50% protein, 5.05% fat, 2.99% ash) for a minimum of 10 days prior to the initiation of the experimental trials. Feed and water were available *ad libitum* throughout the study and photoperiod was determined by natural lighting.

Test feeds consisted of the complete and balanced rodent ration (Bio-Serv) to which 0.00%, 0.50%, 0.75%, 1.00%, 2.00%, and 5.00% tannic acid were added to the test rations replacing undigestible fiber. The feeding trials lasted 7 days for each of the 6 diets. Six different squirrels were fed diets in sequence from tannic acid-free rations through the 5.00% tannic acid diet. Days 1 through 3 were allowed for the squirrels to adjust to the experimental diets. Daily feed consumption did not shift significantly within trial periods for individual squirrels even though feed was available *ad libitum*. Days 4 through 7 served as the collection period when feces was saved for digestibility determinations. Uncontaminated feed and feces samples were dried at 80C for 24 hours to determine dry matter content, then ground to pass a 20 mesh screen in a Wiley mill and stored in airtight containers for later analysis. Lipid content of feed and feces was determined by standard Soxhlet extraction procedures using purified ether as the solvent. Results of the feeding trials were statistically analyzed by one way analysis of variance and, if significant, by Duncan's Multiple Range Test. The 0.05 level of significance was applied.

Results and Discussion

Average daily consumption rates were highest for the control diet and lowest for the 0.75% tannic acid feed (Table 1). Differences in consumption rates, ex-

TABLE 1. *Mean (± Standard Deviation) daily consumption rates of red squirrels feeding on diets with variable tannic acid content.*

	Consumption			
	g-feed/day		per 100g BW/day	
Diet	$\bar{X}$	± SD	$\bar{X}$	± SD
0.00% Tannic Acid	16.29	1.01	8.82	0.23
0.50% Tannic Acid	12.15	0.93	6.58	0.67
0.75% Tannic Acid	11.23	0.57	6.08	0.59
1.00% Tannic Acid	12.48	1.12	6.76	0.67
2.00% Tannic Acid	12.17	0.87	6.60	0.48
5.00% Tannic Acid	12.75	1.02	7.45	0.93

pressed either as per squirrel or per 100 g body weight, among diets were insignificant except for the control diet which was significantly higher than each tannic acid feed. Although consumption data for red squirrels is lacking in the literature, these red squirrels consumed even the least preferred ration at a rate from 31.7% to 63.7% higher than gray or fox squirrels consuming native forages and commercial rations (data recalculated from:6,14,17,42) on a per body weight basis. Since consumption rates for boreal squirrels are apparently linked directly with metabolized nutrient efficiency (14), the high feed consumption observed in this study could be expected for an artificially balanced ration. Additionally, the increased metabolic rate for the smaller red squirrel could result in at least a portion of this increased overall consumption rate. The observed reduction in relative consumption of feeds containing any concentration of tannic acid is at least partially consistent with the inverse relationship of tannin content and food preferences by fox squirrels described by Ofcarcik et al (35). Conversely Smith and Follmer (44) concluded that tannin content is not an important factor in food preferences by squirrels. The data presented in this study suggests that intake is negatively affected by tannic acid content at any level, however, increased dietary concentration of this particular phenolic is not matched by a proportional (or even more extensive) reduction in intake. The suggestion by these data that reduced intake represents a qualitative rather than quantitative phenomenon must be carefully tempered by the fact that the inclusion of only one phenolic compound in the diet of this study may considerably underestimate the complexity of relationships observed with native forages.

Dry matter digestibility of the control ration averaged 92.57% and ranged from 91.44% to 94.49% (Figure 1). The affect of tannic acid on apparent dry matter digestibility was insignificant for diets containing 0.75, 1.00, 2.00, and 5.00% tannic acid, but was significantly reduced for the 0.50% tannic acid which averaged 90.57% dry matter digestibility. These data are contrary to the pattern that increased phenolic content is equated with reduced digestibility as found in a variety of other herbivores including avians (31), rodents (49), and ungulates (29). That digestibility was most affected in the 0.50% tannic acid ration is likely a result of the sequential feeding schedule in which it was the first phenolic containing diet offered. Apparently, red squirrels require an extended adaptation period (greater than the 3 day pretrial period) to adjust to the ingested tannic acid. Subsequent consumption of tannic acid is more easily tolerated since its adverse effects on digestion are clearly neutralized. Cecal digestors (including red squirrels) possess an active array of gut microflora which may respond to dietary phenolics. Rumen microorganisms are known to respond in terms of relative species abundance to dietary tannin content and exhibit no apparent deleterious effects upon the host by tannins comprising up to 2.5% of dietary intake (4). This adaptation by these red squirrels could also involve, in whole or in part, adjustments in the enzymatic activity and/or secretion rate by the squirrels.

The increased apparent dry matter digestibility (although not statistically significant) in the 0.75% and 5.00% diets over the control diet (Figure 1) suggests that tannic acid itself may be digestible since it replaced, as a percentage, undigestible fiber in the rations. Although the extent of tannic acid digestibility cannot be determined by this study, if indeed it is assimilated by red squirrels, there exists a potential for extensive histological and metabolic pathology. Circulating tannic acid has been demonstrated to precipitate acute hepato-toxicity in rats (36), reduced fundicity and result in renal abnormalities (24).

Predictably, lipid digestibility was high for the artificial rations fed in this study (Figure 1). Statistical deviation among diets was observed, but only in the

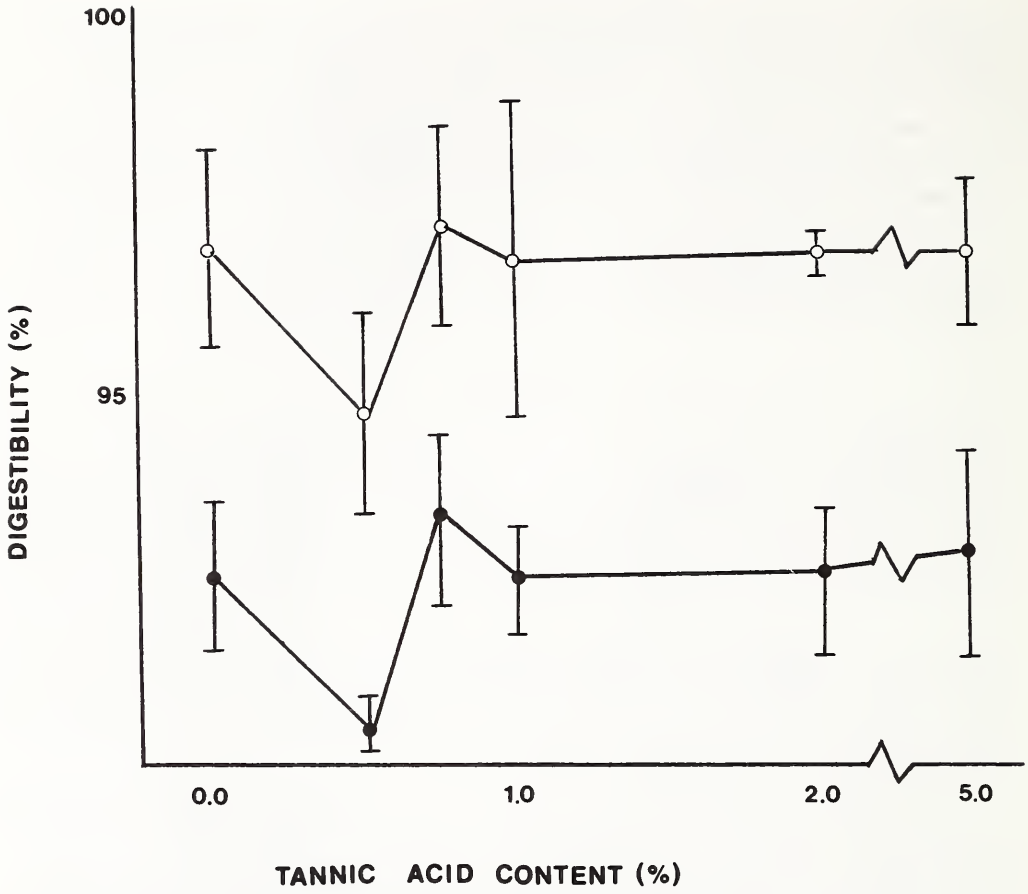

FIGURE 1. *Dry matter (closed circles) and lipid (open circles) digestibility by red squirrels on artificial diets with variable tannic acid content. Bars indicate ± 1 standard deviation.*

0.50% tannic acid diet, as was the case in dry matter digestibility. This too, indicates that red squirrels or their gut floras require an extended adaptation period to physiologically adjust to dietary tannic acid. This is especially noteworthy because the effects of phenolics on dietary lipid digestion have been generally ignored, probably because phenolics are not known to form complexes with nonpolar entities while complexes are readily formed with proteins and carbohydrates (24). That phenolics reduce protein and dry matter digestibility has been well documented, however, the actual site(s) of phenolic involvement is unknown. Due to the multitude of potential binding substrates available in the digestive system, phenolics could complex with proteins and carbohydrates from dietary, secretory, or epithelial sources. The observed effect on lipid digestibility during the adaptation to tannic acid would indicate the extensive involvement of complex formations with endogenous sources since complexes with dietary fats is unlikely. Fecal microflora of these squirrels was apparently altered by dietary tannic acid (10). However, alterations of gastric and intestinal secretions or changes in absorptive epithelium may be more important since lipid digestion is generally attributed to the foregut of cecal digestors (20).

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Quaternary Records of the Pygmy and Smoky Shrews from South-central Indiana Caves

RONALD L. RICHARDS, Indiana State Museum
202 North Alabama Street, Indianapolis, Indiana 46204

Both the Pygmy (*Microsorex hoyi*) and the Smoky (*Sorex fumeus*) Shrews were just recently trapped in south-central Indiana (7; John O. Whitaker, Jr., personal communication, November, 1982). Lyon (1936) had noted that the Pygmy Shrew might possibly occur in Indiana, anywhere in the state, and that the Smoky Shrew might possibly occur in the northern counties as well as around the cold cave entrances in southern Indiana (23). Hall and Kelson (1959) and Burt and Grossenheider (1964) had correctly indicated the presently known range of the Smoky Shrew in Indiana, though without any specimens for documentation (19; 5). Mumford (1969) made no mention of either shrew (24). The only reference to fossil and sub-Recent shrews in Indiana was in 1972 (29). Further identification of those and additional shrew remains form the basis of this paper.

Identifications were with Guilday's "Keys" to the genera and species of Eastern North American soricids based on lower jaws (11), and with comparative material donated and on loan from the Department of Life Sciences, Indiana State University, and material examined at the Museum, Michigan State University, and the Field Museum of Natural History, Chicago. Comparative measurements, made with an ocular micrometer at 15X, are illustrated in Figure 1. The cave remains, generally from undescribed faunas, are temporarily on file with the author. Abbreviations: L, left; R, right; i, p, m, lower incisor, premolar (unicuspid), and molar, respectively; P, M, upper premolar and molar; $\overline{X}$, Mean; O.R., Observed Range; N, Number of individuals; B.P., Before Present (1950 A.D.); mm., millimeters.

Remains of 8 individuals of *Microsorex hoyi* (4L, 6R dentaries; 2 palates; 1R maxilla) were recovered from Freeman's Pit, northwest Monroe county, Indiana (Figure 2). Six of the shrews occurred with an abundant microfauna in the top 11 inches of a 24 inch laminated silt/clay deposit at the bottom of the 97 foot pit. Palates of two individuals were from nearby deposits. A single radiocarbon date on bone from the lowermost level of *Microsorex* occurrence in the laminated deposit (Level SL-1) gave a date of 2,315 $\pm$ 65 years B.P. Remains of *Microsorex* showed no difference in size or morphology throughout the laminated deposit.

Microsorex has been found fossil in 14 sites of 10 other states (Figure 3). *Microsorex pratensis* has been reported from the Middle Pleistocene of Kansas (20), and *M. minutus* from the Late Pleistocene of Arkansas (3). All other fossils are of *M. hoyi*. If the small *M. h. thompsoni* and *M. h. winnemana* of the Appalachians are to be regarded as a separate species, *M. h. thompsoni* (22), then most of the eastern North American cave fossils should be placed in that taxon. While several of the sites appear to be out of the modern range of *Microsorex* (eg. Crankshaft Cave, Missouri; Meyer Cave, Illinois; Freeman's Pit, Indiana; Robinson Cave, Tennessee; Peccary Cave, Arkansas and Little Box Elder Cave, Wyoming), the rarity of the Pygmy shrew and its often sporadically recovered specimens makes its modern distribution difficult to determine. It is apparent, however, that it was more common and widespread in the east and had extended its range further south in the Late Pleistocene (14; 16).

Microsorex exhibits a Bergmann's Response, with the largest individuals in Alaska and Canada, and the smallest in the Rocky and Appalachian Mountains

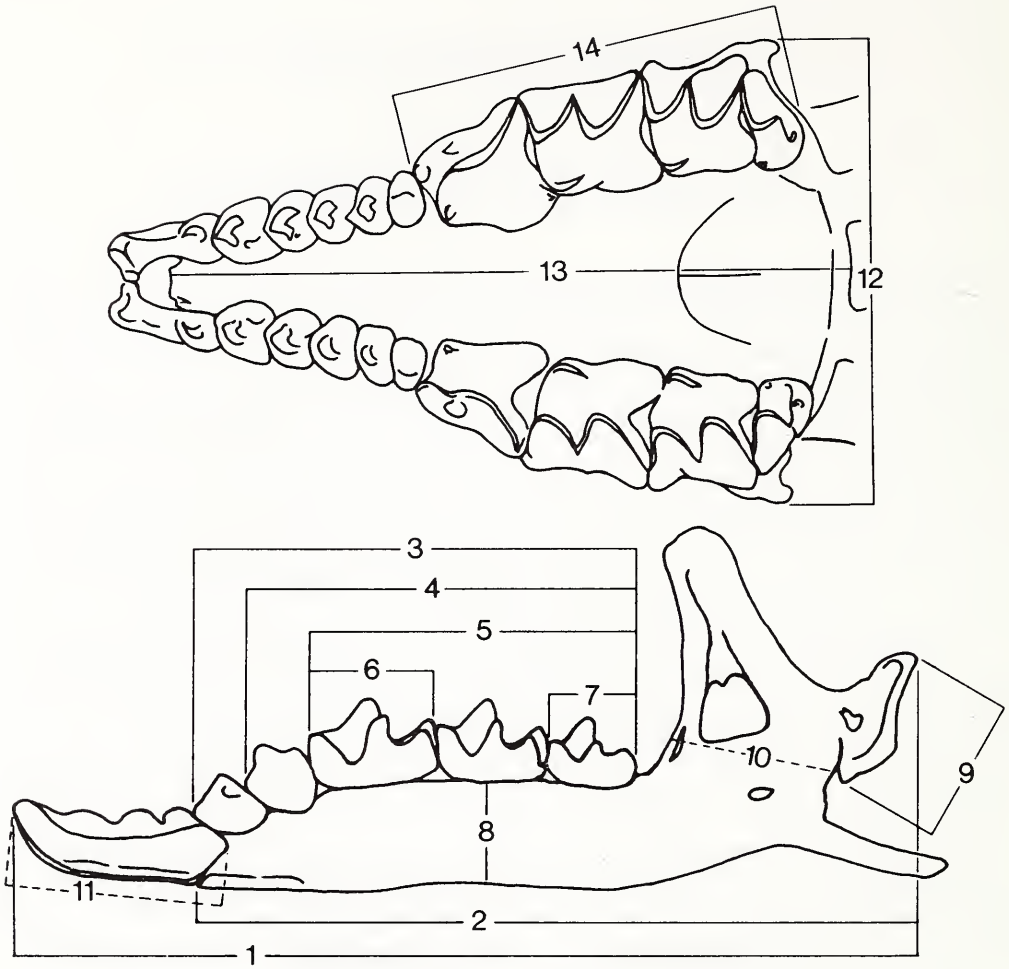

FIGURE 1. *Palate (above) and right dentary (below) of Sorex fumeus, illustrating comparative measurements. 1, Total length of mandible (including incisor); 2, Total length of dentary; 3, p3-m3; 4, p4-m3; 5, m1-m3; 6, Length, m1; 7, Length, m3; 8, Depth of dentary at m2; 9, Width of condyle; 10, Width (least) of vertical ramus; 11, Length of incisor; 12, Maxillary width; 13, Palatal length; 14, P4-M3. Note, dentary lays flat on labial side for measuring. Measurements with dotted lines are taken from the opposite side.*

(22). Most of the eastern North American fossils (eg. from New Paris #4, Pa.; Bootlegger Sink, Pa.; Clark's Cave, Va.; Robinson Cave, Tn. and Welsh Cave, Ky.) have the small size of *M. h. thompsoni/winnemana* in the Appalachians today (17). Absent were bones of the larger northern *Microsorex* perhaps to have been anticipated from late glacial age sites. The Pygmy shrews from Freeman's Pit were small (Table 1; Figure 4). They do have some size overlap with the smallest individuals from Robinson and Baker Bluff Caves, Tennessee.

Long (22) suggested that the Pleistocene glaciations displaced *Microsorex* populations into refugia, with the warm Recent epoch restricting some to mountain ranges. The several subspecies would have developed from the isolated populations, with *M. h. hoyi* ranging into the region formerly covered by the Wisconsinan glacial ice. Guilday et. al. (17) suggest the possibility that *M. hoyi* may have spread into eastern Canada from the west during postglacial times. They note that "except for the Gaspé Peninsula, where it is replaced by *M. hoyi*, the nor-

FIGURE 2. *Locations of Indiana caves containing shrew species discussed in text. Microsorex hoyi: F, Freeman's Pit, Monroe Co.; Sorex fumeus: H, Hidden Pit Cave, Owen Co.; F, Freeman's Pit, Monroe Co.; S, Showcase Cave, Monroe Co.; A, Anderson Pit Cave, Monroe Co.; K, King Leo Cave, Harrison Co.; D, Devil's Staircase Cave, Harrison Co.; J, N. Jim Cave, Harrison Co.; R, Owl Roost deposit, Harrison Co.; Misc. shrew faunas: 1, Sullivan's Cave, Lawrence Co.; 2, Carcass Crypt Cave, Lawrence Co.; 3, Gibbs Pit, Harrison Co..*

thern limit of *M. thompsoni* appears to be defined by the Great Lakes—St. Lawrence waterway, which may have acted as a barrier to its northward spread in postglacial times". The Freeman's Pit *Microsorex* also suggest that the larger *M. h. hoyi* had been absent in eastern North America from the Late Pleistocene and thereafter, suggesting, as Guilday et. al. had pointed out, that *M. hoyi* and *M. thompsoni* were at one time geographically separated populations, arguing for specific distinction. To date, the Freeman's Pit bones represent the smallest Pygmy Shrews known, and appear to have genetic affinities with the populations inhabiting the southern Appalachians today.

Sorex fumeus, the Smoky Shrew, is documented in Indiana by fossil and Recent age specimens representing about 47 individuals (30L, 45R dentaries; 10 palates; 9L, 8R maxillae) from 7 caves in Owen, Monroe and Harrison counties (Figure 2). One palate was recovered from an "old" owl pellet deposit on a Harrison county bluff. Lack of *S. fumeus* bones from caves of interjoining counties is probably due to insufficient collecting. In the two Indiana caves where the deposits were excavatable in sequential units (Freeman's Pit, Monroe Co. and N. Jim Cave, Harrison Co.) the lowermost levels produced larger *S. fumeus* den-

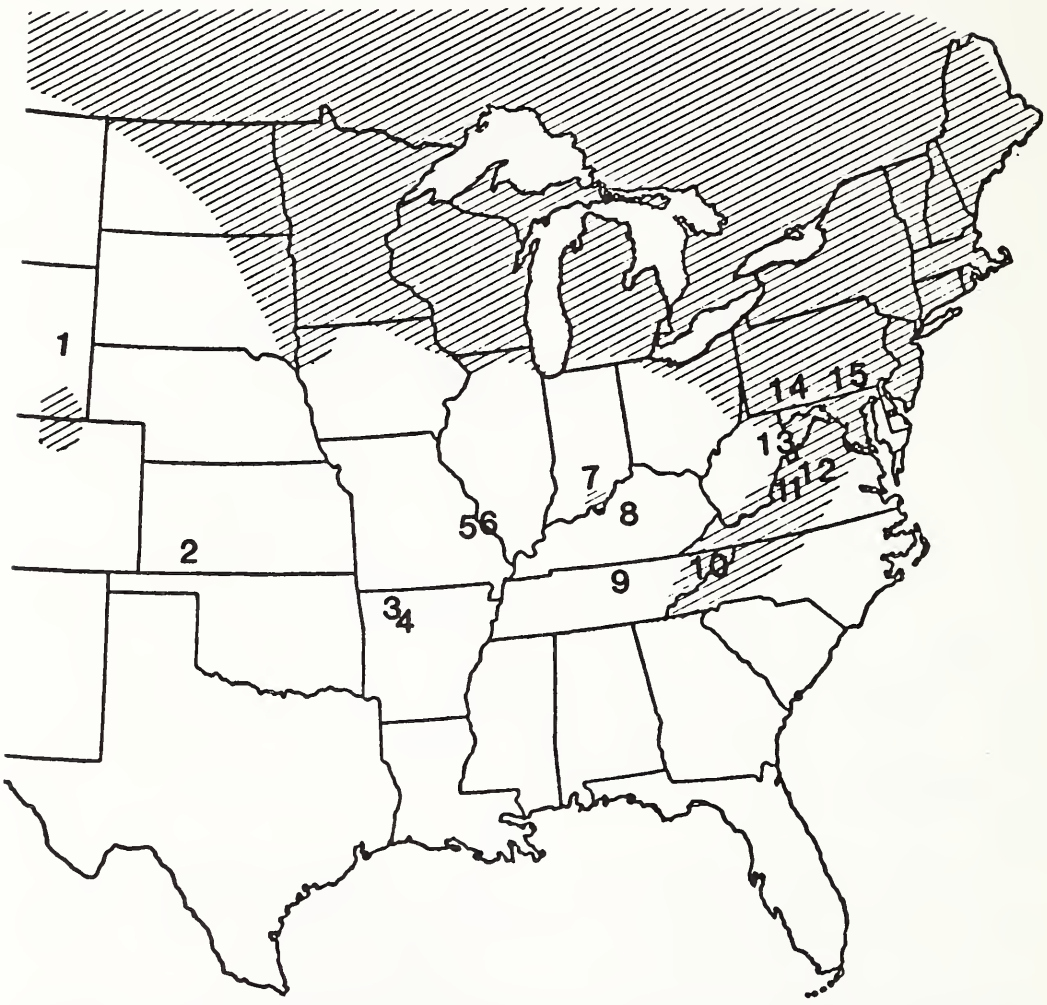

FIGURE 3. *Recent and fossil range of Microsorex. Hatched area: modern range (data modified from Hall and Kelson, 1959, Long, 1972 and Caldwell, 1980). Fossil sites (and literature citation): 1, Little Box Elder Cave, Converse Co., Wyo. (1); 2, M. pratensis, Meade Co., Kan. (20); 3, M. minutus, Conard Fissure, Newton Co., Ark. (3); 4, Peccary Cave, Newton Co., Ark. (9); 5, Crankshaft Cave, Jefferson Co., Mo. (27); 6, Meyer Cave, Monroe Co., Il. (26); 7, Freeman's Pit, Monroe Co., In.; 8, Welsh Cave, Woodford Co., Ky. (15); 9, Robinson Cave, Overton Co., Tn. (14); 10, Baker Bluff Cave, Sullivan Co., Tn. (18); 11, Clark's Cave, Bath Co., Va. (17); 12, Natural Chimneys, Augusta Co., Va. (11); 13, Eagle Cave, Pendleton Co., W. Va. (12); 14, New Paris #4, Bedford Co., Pa. (16); 15, Bootlegger Sink, York Co., Pa. (13).*

taries and teeth (Figures 5 and 6). The change is gradual in Freeman's Pit, perhaps because of the number of excavation units and/or a continuous deposition, and quite abrupt in N. Jim Cave, with only a sparse representation of the smaller *S. fumeus*.

The great discrepancy in size of the large N. Jim Cave *S. fumeus* from all other Indiana cave specimens (Figure 5 and Table 2) made their identification at first questionable, since they ranged into the large size of *Sorex palustris*, the Water Shrew and *S. arcticus*, the Arctic Shrew. Characters of the ml (used by Guilday) to separate these species appeared intermediate in some instances. Scrutiny of the lower incisor, however, allowed separation of 64 of 67 modern

TABLE 1. *Measurements of Microsorex, Freeman's Pit, Monroe Co., Indiana (mm).*

Measurement	$\bar{X}$	O.R.	N (of elements)
Total length of mandible (including i):	7.38	7.20-7.57	8
Total length of dentary:	5.95	5.81-6.10	9
p3-m3:	3.50	3.43-3.60	4
p4-m3	3.19	3.13-3.25	5
m1-m3:	2.64	2.50-2.76	9
Length, m1:	1.04	0.95-1.09	10
Length, m3:	0.72	0.60-0.77	9
Depth of dentary at m2:	0.88	0.80-0.97	10
Width of condyle:	1.36	1.30-1.40	8
Height of vertical ramus:	2.90	2.78-3.07	9
Maxillary width:	3.75	3.70-3.80	2
Palatal length:	4.71	4.61-4.8	2
P4-M3:	3.07	2.97-3.17	2

comparative specimens (*S. palustris*: British Columbia, 1; Ontario, 3; Maine, 2; Vermont, 1; Wisconsin, 5; Wyoming, 1; Colorado, 3; California, 10; *S. arcticus*: Alaska, 2; Canada, 2; Minnesota, 2; Wisconsin, 7; *S. fumeus*: Quebec, 1; Maine, 2; New Brunswick, 1; New York, 1; West Virginia, 4; North Carolina, 1; Indiana (modern), 18). Incisors of 3 specimens from Harrison Co., In. were too worn to make a *S. fumeus/palustris* distinction. In *S. palustris* and *S. fumeus* a ridge extends along the inner inferior edge of the lower incisor: it rises gradually upward at

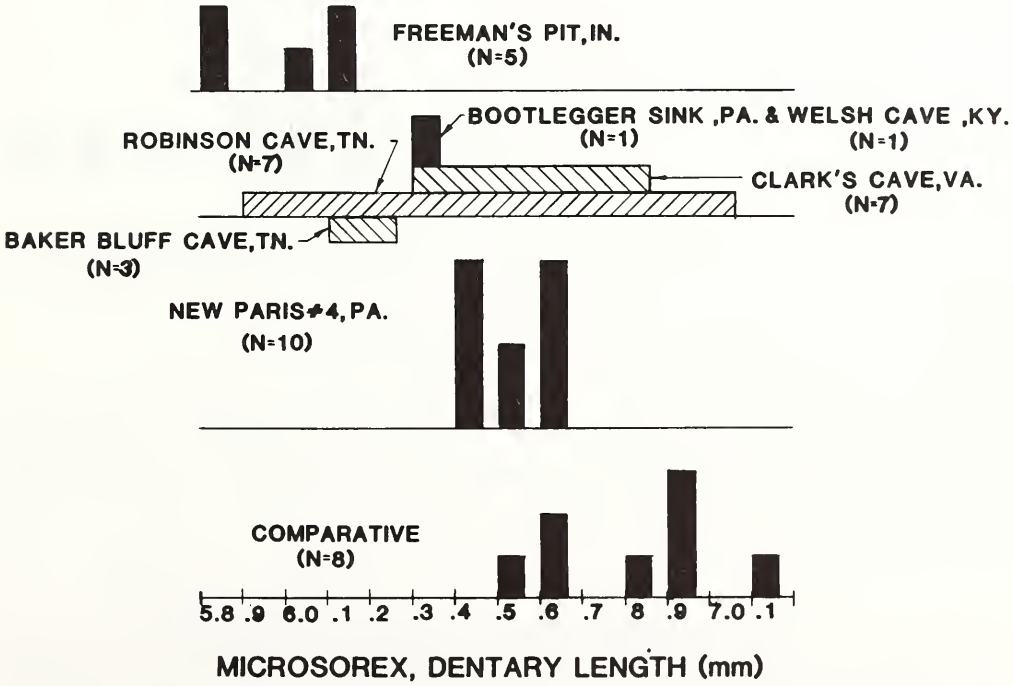

FIGURE 4. *Variation of dentary length in Microsorex, eastern North American sites. Black vertical bars, individual measurements. N= number of individuals. Hatched horizontal bars, encompass the Observed Range (individual measurements not available). Modern comparative specimens, left to right: Alberta; New York/Pennsylvania (New Paris #3, Guilday, et. al., 1977); Wisconsin; Illinois/Wisconsin/Maine; Wisconsin.*

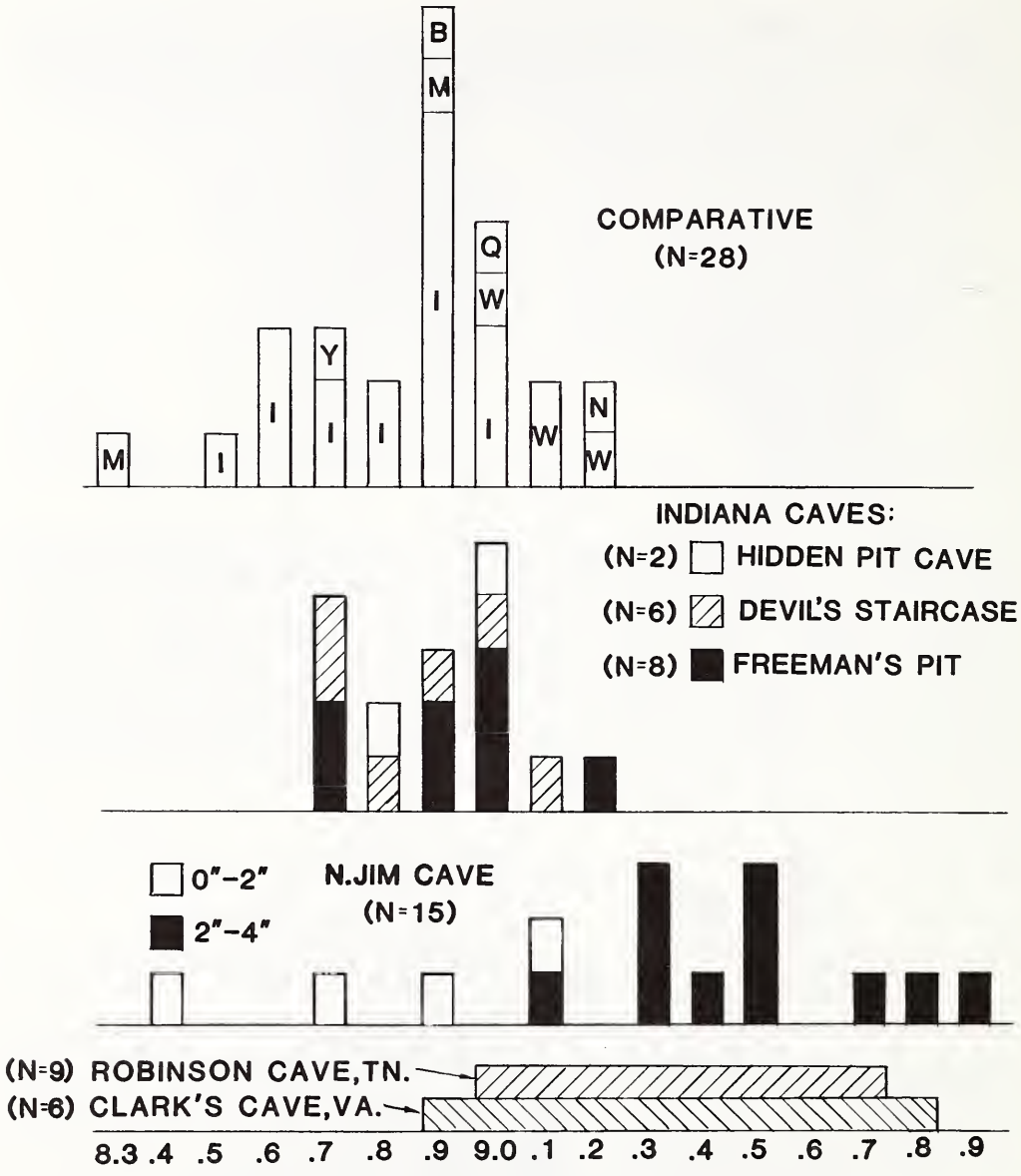

FIGURE 5. *Sorex fumeus, variation in dentary length. Vertical bars, individual measurements. N= number of dentaries. Horizontal bars, encompass the Observed Range (individual measurements not available). Modern comparative specimens: B, New Brunswick, 1; I, Indiana (Harrison/Crawford Cos.), 18; M, Maine, 2; N, North Carolina, 1; Q, Quebec, 1; W, West Virginia, 4; Y, New York, 1.*

the tip of the tooth in *palustsris*, but bows strongly upward in *fumeus*. In *S. arcticus* the ridge extends nearly along the midline of the tooth. Guilday's use of the postmandibular foramen in identifying *S. arcticus* was supported. By lower incisor criteria, all medium and large size Indiana cave *Sorex* in this study appear to represent *Sorex fumeus*.

The Smoky Shrew has been found fossil on 9 sites of six other states (Figure 7). The earliest record is from the Conard Fissure deposit, Arkansas (3). Relying on relatively mature forest cover, *S. fumeus* is presently the most common *Sorex*

TABLE 2. *Measurements of Indiana Sorex fumeus, modern and cave specimens (mm.).*

Site	$\bar{X}$	O.R.	N (of elements)
Total length of mandible (including incisor):			
Modern, Harrison/Crawford Cos.:	10.82	10.49-11.08	18
Freeman's Pit, Monroe Co.:			
Laminated deposit: Level I:	10.91	10.74-11.01	3
Level II:	11.13	— —	1
Level SL-1:	11.26	— —	1
Other deposits: "Chimney":	10.74	— —	1
"Attic, Room #4":	11.26	— —	1
Hidden Pit Cave, Owen Co.:	11.0	10.99-11.0	2
Devil's Staircase Cave, Harrison Co.:	10.95	10.81-11.12	5
N. Jim Cave, Harrison Co.: 0-2":	11.0	10.63-11.5	4
2-4":	11.79	11.42-12.38	10
Total length of dentary:			
Modern, Harrison/Crawford Cos.:	8.80	8.37-9.04	18
Freeman's Pit, Monroe Co.:			
Laminated deposit: Level I:	8.92	8.67-9.18	4
Level II:	8.92	— —	1
Level SL-1:	9.0	— —	1
Other deposits: "Chimney":	8.67	— —	1
"Attic, Room #4":	9.0	— —	1
Hidden Pit Cave, Owen Co.:	8.89	8.78-9.0	2
Devil's Staircase Cave, Harrison Co.:	8.86	8.68-9.911	6
N. Jim Cave, Harrison Co.: 0-2":	8.77	8.41-9.11	4
2-4":	9.48	9.13-9.88	11
p3-m3:			
Modern, Harrison/Crawford Cos.:	5.19	5.01-5.32	18
Freeman's Pit, Monroe Co.:			
Laminated deposit: Level I:	5.26	5.20-5.37	3
Level II:	5.27	— —	1
Hidden Pit Cave, Owen Co.:	5.20	— —	1
Devil's Staircase Cave, Harrison Co.:	5.30	5.26-5.33	2
N. Jim Cave, Harrison Co.: 0-2":	5.37	5.10-5.54	3
2-4":	5.39	5.02-5.60	4
p4-m3:			
Modern, Harrison/Crawford Cos.:	4.58	4.41-4.75	18
Freeman's Pit, Monroe Co.:			
Laminated deposit: Level I:	4.61	4.54-4.71	5
Level I&II:	4.68	— —	1
Level II:	4.63	— —	1
Other deposits: "Attic, Room #4":	4.57	— —	1
Hidden Pit Cave, Owen Co.:	4.60	4.51-4.68	2
Devil's Staircase Cave, Harrison Co.:	4.67	4.63-4.72	3
N. Jim Cave, Harrison Co.: 0-2":	4.71	4.49-5.01	4
2-4":	4.73	4.41-4.90	4
m1-m3:			
Modern, Harrison/Crawford Cos.:	3.74	3.60-3.90	18
Freeman's Pit, Monroe Co.:			
Laminated deposit: Level I:	3.77	3.70-3.81	5
Level I&II:	3.77	— —	1
Level II:	3.76	— —	1
Level SL-1:	4.02	— —	1
Other deposits: "Chimney":	3.76	— —	1
"Attic, Room #4":	3.80	— —	1
Hidden Pit Cave, Owen Co.:	3.80	3.74-3.86	2
Devil's Staircase Cave, Harrison Co.:	3.82	3.76-3.90	5
N. Jim Cave, Harrison Co.: 0-2":	3.89	3.68-4.14	4
2-4":	3.95	3.63-4.07	7

TABLE 2.—Continued

Site	$\bar{X}$	O.R.	N (of elements)
Length, m1:			
Modern, Harrison/Crawford Cos.:	1.47	1.41-1.54	18
Freeman's Pit, Monroe Co.:			
Laminated deposit: Level I:	1.49	1.43-1.54	7
Level I&II:	1.51	1.48-1.56	3
Level II:	1.54	1.50-1.57	2
Level SL-1:	1.61	1.60-1.61	2
Other deposits: "Chimney":	1.52	1.50-1.55	3
"Attic, Room #4":	1.47	— —	1
Hidden Pit Cave, Owen Co.:	1.50	— —	2
Showcase Cave, Monroe Co.:	1.50	— —	1
Anderson Pit Cave, Monroe Co.:			
Woodrat Den #1: Upper Level:	1.50	— —	1
Deposit 13.2	1.58	— —	1
King Leo Cave, Harrison Co.:	1.50	— —	1
Devil's Staircase Cave, Harrison Co.:	1.52	1.45-1.57	5
N. Jim Cave, Harrison Co.: 0-2"	1.55	1.45-1.65	4
2-4":	1.60	1.48-1.66	23
Maxillary width:			
Modern, Harrison/Crawford Cos.:	5.21	5.00-5.47	17
Owl Roost Deposit, Harrison Co.:	5.20	— —	1
Devil's Staircase Cave, Harrison Co.:	5.20	— —	1
N. Jim Cave, Harrison Co.: 2-4"	5.54	5.48-5.6+	2
Palatal length:			
Modern, Harrison/Crawford Cos.:	7.59	7.30-7.83	17
Owl Roost Deposit, Harrison Co.:	7.54	— —	1
Devil's Staircase Cave, Harrison Co.:	7.58	— —	1
N. Jim Cave, Harrison Co.: 0-2":	8.10	— —	1
2-4":	8.42	8.30-8.54	2
P4-M3:			
Modern, Harrison/Crawford Cos.:	4.55	4.36-4.73	18
Freeman's Pit, Monroe Co.:			
Laminated deposit: Level I&II:	4.70	— —	1
Owl Roost Deposit, Harrison Co.:	4.70	— —	1
Devil's Staircase Cave, Harrison Co.:	4.50	— —	1
N. Jim Cave, Harrison Co.: 2"-4":	4.87	4.77-5.05	3

in the central and southern Appalachians; it was relatively scarce, however, on most mid-Appalachian faunal sites, perhaps indicating more open woodlands in the Late Pleistocene than at present (17).

Sorex fumeus appears to exhibit a reverse Bergmann's Response, the southern populations (eg. West Virginia) having larger individuals than those to the north; there is a general conformity with the cave fossils (Figure 5). Dentaries from N. Jim Cave, Indiana have the large size of the fossils from Clark's Cave, Va. and Robinson Cave, Tn.

The Indiana data suggests that there was a change to a smaller *S. fumeus* in more recent periods. Perhaps this size change can be used in correlating at least local faunal deposits. If so, the N. Jim Cave bones would be older than those from Freeman's Pit, among others. If the clinally variable *S. fumeus* populations did adjust their ranges to the rigorous glacial climates of the Pleistocene, perhaps the size of *S. fumeus* (as well as *Microsoreq*) fossils could be used in paleoclimatic interpretation (eg. larger *S. fumeus*, warmer climates), as has been done with *Blarina brevicauda*, the Short-tailed Shrew (10). More C-14 dated, stratigraphically excavated, shrew faunas are needed from the Midwest.

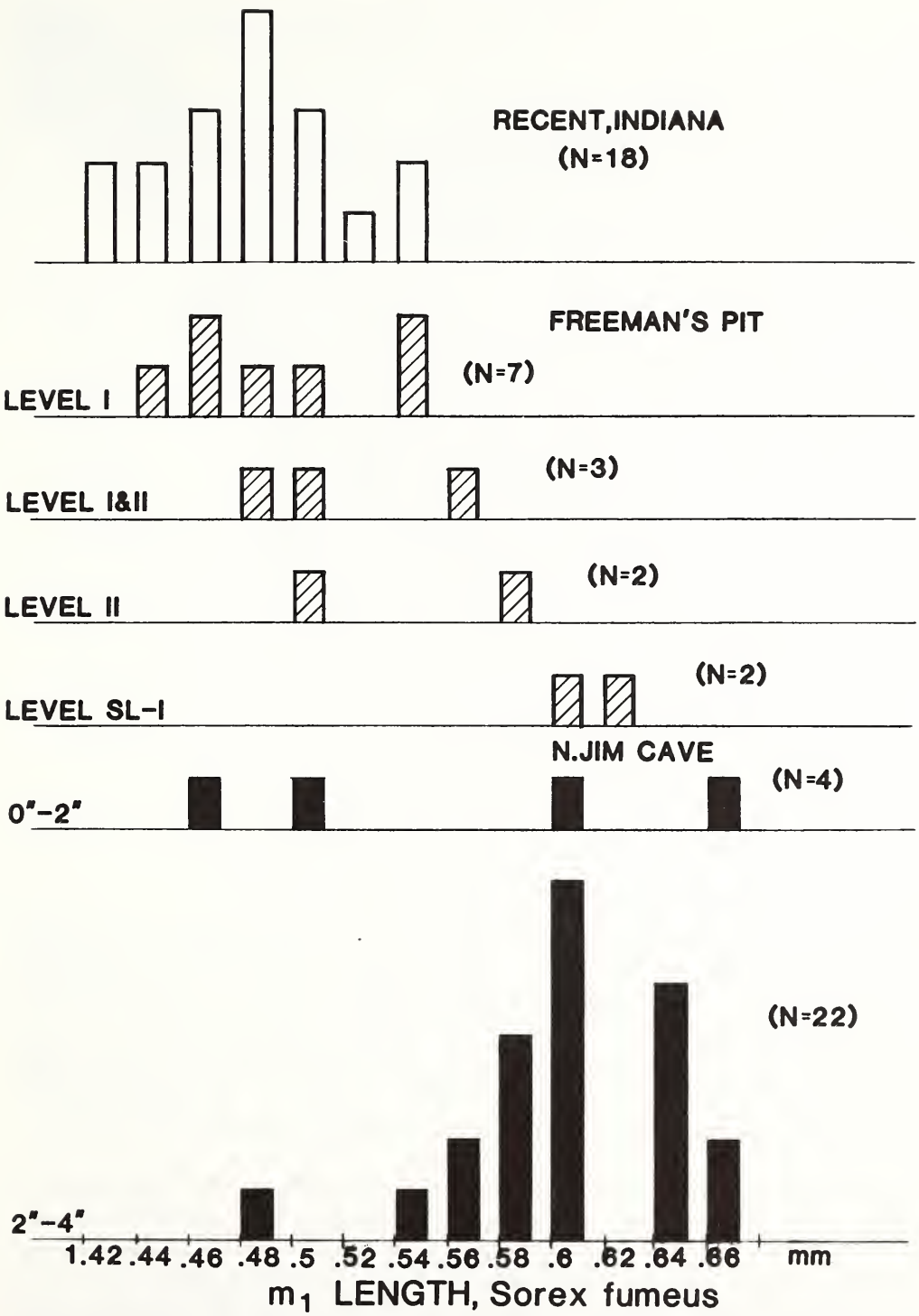

FIGURE 6. *Indiana Sorex fumeus, variation in m1 length. Vertical bars, individual measurements. N= number of m1's measured. Recent, Indiana from Harrison/Crawford counties.*

To put the abundance of *Microsorex hoyi* and *Sorex fumeus* in perspective, total shrew faunas from several Indiana caves are listed (Table 3). *Blarina brevicauda*, the Short-tailed Shrew, one of Indiana's most abundant mammals, usually dominated the shrew component of a cave fauna. A dozen other cave

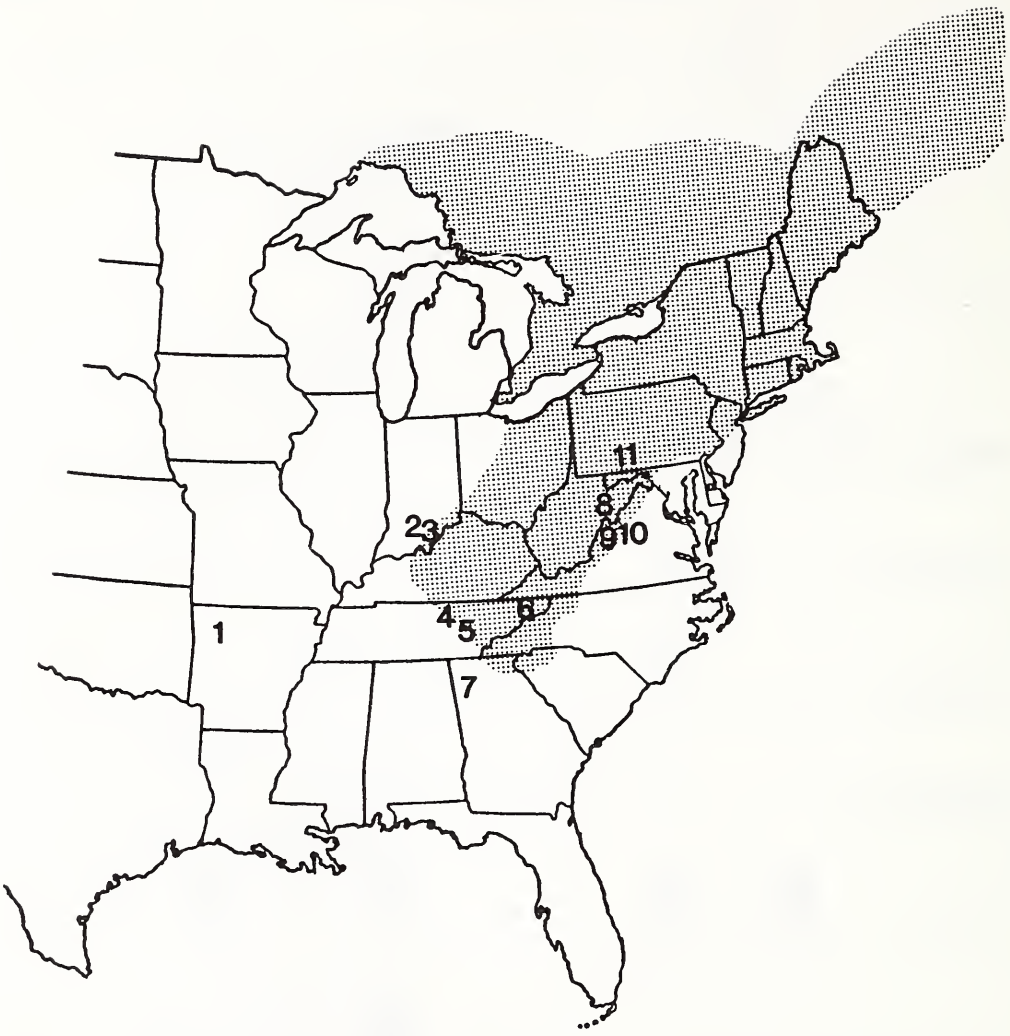

FIGURE 7. *Recent and fossil range of Sorex fumeus. Stippled area: modern range (after Hall and Kelson, 1959). Fossil sites (with literature citation): 1, Conard Fissure, Newton Co., Ark. (3); 2,3 Owen/Monroe and Harrison Co., In. sites (see Fig. 2); 4, Robinson Cave, Overton Co., Tn. (14); 5, Banshee Hole, Cumberland Co., Tn. (8); 6, Baker Bluff Cave, Sullivan Co., Tn. (18); 7, Ladds, Bartow Co., Ga. (28); 8, Eagle Cave, Pendleton Co., W. Va. (12); 9, Clark's Cave, Bath Co., Va. (17); 10, Natural Chimneys, Augusta Co., Va. (11); 11, New Paris #4, Bedford Co., Pa. (16).*

deposits, not listed in Table 3, contained *B. brevicauda* as the only shrew. It is presently found in most habitats except those that are excessively wet (24). *Cryptotis parva*, the Least Shrew, was usually absent, or represented less than 8.0% of a shrew fauna. In the Harrison county owl roost deposit, however, this grassland-inhabitating species accounted for 38.0% of the shrews, reflecting its availability for owl predation. *Sorex fumeus* inhabits moist woodlands where rocks, fallen trees, or brush provide good ground cover (2). It consistently represented 5.0-7.0% of the shrews throughout the upper four levels of Freeman's Pit, Monroe county. In extreme south-central Indiana, where the shrew has been recently trapped, the percent was much higher: 33.0% in Devil's Staircase Cave, Harrison county, and 79.0% in N. Jim Cave, Harrison County, where the predominantly large version of *S. fumeus* outnumbered *B. brevicauda* 5 to 1. It was nearly absent from the "old" owl roost deposit of the same county. *Microsorex*, often found in a wooded

TABLE 3. *Indiana Cave Deposit Shrew Faunas*[1]

Site	Blarina brevicauda	Cryptotis parva	Microsorex hoyi	Sorex fumeus	Sorex longirostris	Sorex cinereus	Sorex cinereus/ longirostris
			Owen County				
Hidden Pit Cave	2 (40%)	— —	— —	3 (60%)	— —	— —	— —
			Monroe County				
Freeman's Pit:							
Laminated sequence:[2]							
Level I:	46 (72%)	1 (2%)	3 (5%)	4 (6%)	10 (16%)	— —	— —
Level I & II:	27 (69%)	2 (5%)	— —	2 (5%)	7 (18%)	1 (3%)	— —
Level II:	11 (61%)	1 (6%)	2 (11%)	1 (6%)	2 (11%)	1 (6%)	— —
Level SL-1:	9 (64%)	— —	1 (7%)	1 (7%)	2 (14%)	1 (7%)	— —
Level SL-2	3 (75%)	— —	— —	— —	— —	— —	1 (25%)
Level SL-4:	1 (50%)	— —	— —	— —	— —	— —	1 (50%)
Other Deposits:							
"Chimney":	3 (43%)	— —	1 (14%)	2 (29%)	— —	— —	1 (14%)
"Bank":	7 (70%)	1 (10%)	1 (10%)	— —	1 (10%)	— —	— —
"Attic,Room #4":	5 (63%)	— —	— —	1 (13%)	— —	1 (13%)	1 (13%)
Showcase Cave	11 (92%)	— —	— —	1 (8%)	— —	— —	— —
Anderson Pit Cave:							
Woodrat Den #1:[3]							
Upper Level:	33 (89%)	— —	— —	2 (5%)	— —	2 (5%)	— —
Lower Level:	12 (86%)	— —	— —	— —	— —	2 (14%)	— —
Deposit 13.1:	7 (88%)	— —	— —	1 (13%)	— —	— —	— —
Deposit 13.2:	4 (80%)	— —	— —	1 (20%)	— —	— —	— —
"Ledge #1":	2 (67%)	— —	— —	— —	1 (33%)	— —	— —
"Ledge #2":	16 (94%)	— —	— —	— —	— —	1 (6%)	— —
			Lawrence County				
Sullivan's Cave:	34 (100%)	— —	— —	— —	— —	— —	— —
Carcass Crypt Cave:	2 (33%)	2 (33%)	— —	— —	2 (33%)	— —	— —
			Harrison County				
King Leo Cave:	34 (94%)	1 (3%)	— —	1 (3%)	— —	— —	— —
Gibbs Pit:	2 (67%)	— —	— —	— —	1 (33%)	— —	— —
Devil's Staircase Cave:	7 (58%)	— —	— —	4 (33%)	— —	1 (8%)	— —
N. Jim Cave:[4]							
0-2":	1 (25%)	— —	— —	3 (75%)	— —	— —	— —
2-4":	3 (12%)	2 (8%)	— —	20 (80%)	— —	— —	— —
Owl Roost Deposit:	49 (58%)	32 (38%)	— —	1 (1%)	2 (2%)	— —	— —

1, The shrew component of faunas from predominantly silt/clay deposits in caves are listed. Cave locations given in Figure 2.

2, The 24-inch, laminated silt/clay deposit was arbitrarily divided into 8 units. From uppermost down: Level I, Level I & II, and Level II (each 3-4 inch units); Level SL-1, SL-2, SL-3, SL-4, SL-5 (each 2-3 inch units).

3, A 5-9 inch deep silt/clay deposit on a ledge, arbitrarily divided into Upper and Lower units.

4, A small bone deposit arbitrarily recovered in 2-inch increments: uppermost (0-2") and below (2-4").

area or grass clearing bordering it (4), represented 0-11.0% of the shrews in Freeman's Pit, Monroe county, but was absent from all other cave sites. *Sorex longirostris*, the Southeastern Shrew, was common in south-central Indiana cave faunas. It accounted for 11.0-18.0% of the shrews in Freeman's Pit. Though Rose (30) trapped most of his specimens in oldfields, it does also inhabit forested areas. *Sorex cinereus*, the Masked Shrew, with few records from south-central Indiana (25), was recovered in lesser numbers (0-8.0%) and more sporadically from cave deposits than the Southeastern Shrew. It occurs in a variety of moist habitats (5).

Some authors have had difficulty in separating fragmented fossils of *Sorex cinereus* and *Sorex longirostris* (14; 17). *Sorex longirostris* is slightly smaller than

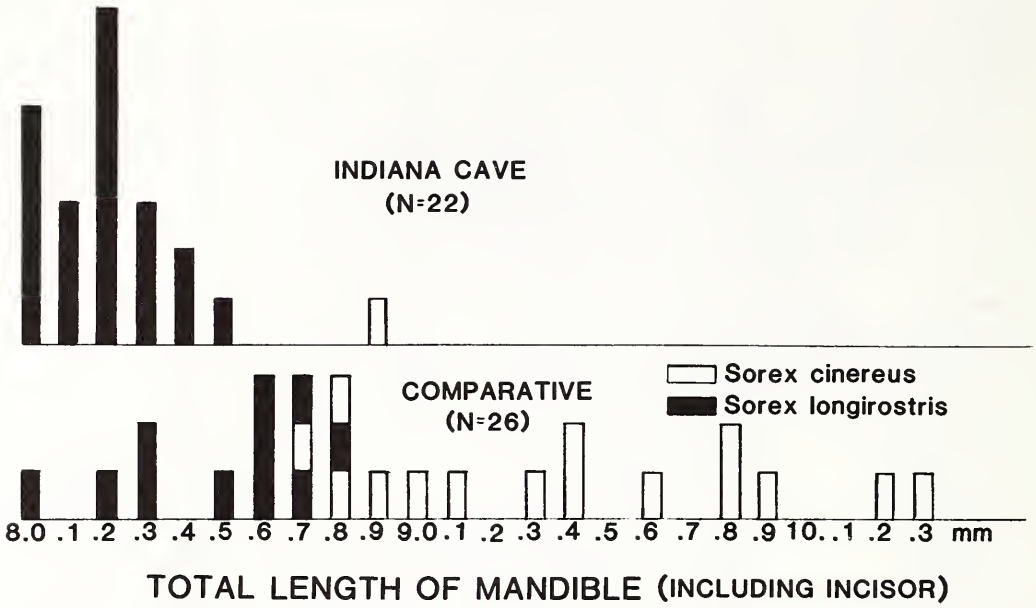
INDIANA CAVE
(N=22)
COMPARATIVE
(N=26)
Sorex cinereus
Sorex longirostris
8.0 .1 .2 .3 .4 .5 .6 .7 .8 .9 9.0 .1 .2 .3 .4 .5 .6 .7 .8 .9 10. .1 .2 .3 mm
TOTAL LENGTH OF MANDIBLE (INCLUDING INCISOR)

FIGURE 8. *Separation of Sorex cinereus and Sorex longirostris by Total length of mandible. Upper, all Indiana cave specimens combined. Lower, modern comparative specimens: Sorex longirostris (black): 8.0, In.; .2, Al.; .3-.5, In.; .6-.8, S. Car. Sorex cinereus (white): .7, Mi.; .8, Mi./In.; .9, In.; 9.0-.1, Mi.; .3-.4, In.; .6-.3, Mi..*

S. cinereus. Both display clinal variation in size. *Sorex cinereus* becomes larger to the north (16), and by the few specimens examined by the author *S. longirostris* appears to get larger to the south. The relative sizes of the 3rd and 4th upper unicuspids, a somewhat unreliable but often used character in separating skulls of the two species (2), can rarely be used since the unicuspids are usually lost in the recovery of fossil material. It was found that any of four measurements of the lower jaw (dentary) will separate most *Sorex* "*cinereus/longirostris*" jaws recovered: Total length of mandible; Total length of dentary; Length of lower incisor, and width of vertical ramus (Figure 1; eg. Figure 8). The latter two measurements were based upon limited material. Measurement of the width of the vertical ramus allows identification of a commonly recovered fragment.

TABLE 4. *Percent of species in eastern North American shrew faunas (no. of individuals).*

Species	New Paris #4, Pa.	Natural Chimneys, Va.	Clark's Cave, Va.	Baker Bluff Cave, Tn.	Robinson Cave, Tn.	Crankshaft Cave, Mo.	Meyer Cave, Il.
Blarina brevicauda	35% (37	42% (56)	42% (97)	54% (149)	36% (83)	86% (663)	20% (262)
Cryptotis parva	– –	4% (5)	– –	1% (3)	2% (5)	.6% (5)	78% (1049)
Microsorex hoyi	10% (11)	5% (7)	3% (7)	3% (9)	7% (17)	1% (10)	–(1)
Sorex arcticus	6% (6)	5% (6)	6% (13)	1% (3)	4% (10)	–(1)	– –
Sorex palustris	1% (1)	3% (4)	3% (7)	– –	1% (2)	2% (12)	– –
Sorex fumeus	1% (1)	8% (10)	4% (10)	8% (22)	16% (37)	– –	– –
Sorex dispar	4% (4)	– –	2% (4)	3% (9)	4% (9)	– –	– –
Sorex cinereus	33% (35)	34% (45)	29% (67)	19% (53)	30% (70)	3% (19)	– –
Sorex longirostris	– –	– –	– –	– –	– –	– –	2% (27)
Sorex sp.	11% (12)	– –	11% (26)	11% (30)	– –	8% (61)	– –
Total No. Individuals	(107)	(133)	(231)	(278)	(233)	(771)	(1339)

The percentages of shrew species from other eastern North American sites are compared in Table 4 *Blarina brevicauda* was usually the dominant shrew (note exception: Meyer Cave, Il.). *Sorex cinereus* was quite abundant within its modern range (New Paris #4, Pa., 33.0%; Clark's Cave, Va., 29.0%; Natural Chimneys, Va., 34.0%), and occasionally out of its modern range (extralocally): Robinson Cave, Tn., 30.0%. *Sorex longirostris*, a southern form, was usually absent on Late Pleistocene sites, even within its modern range (eg. Robinson Cave, Tn. and Clark's Cave, Va.). *Sorex arcticus* was found well south of its present range, in small numbers, on most of the sites. *Sorex palustris* was extralocal in small numbers on several sites. *Microsorex* usually represented under 10.0% in a given fauna. *Sorex fumeus* was usually of a minor percent, even within its modern range, but sometimes occurred in greater numbers when the larger form was represented (Robinson Cave, Tn., 16.0%; N. Jim Cave, In., 79.0%). *Sorex dispar*, an Appalachian form, was occasionally present in small numbers, and was extralocal at Robinson Cave, Tn. The shifting ranges of shrews on most eastern sites was presumably in response to the cool, moist glacial climates.

As more late glacial shrew faunas become known in Indiana the following components may be recognized: dominance of *Blarina brevicauda*; abundance of *Sorex cinereus* with suppressed numbers of *S. longirostris*; *Sorex fumeus* in small to medium numbers; *Microsorex* in small numbers; presence of *Sorex arcticus* and *S. palustris* in small numbers. The large size of some *Sorex fumeus* and small size of *Microsorex* fossils often in deposits of presumed late glacial age, as well as the structure of interglacial shrew faunas of the Midwest need to be further investigated.

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